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Biologic Effects of Atmospheric Pollutants

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FLUORIDES

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FLUORIDES

Committee on
Biologic Effects of
Atmospheric Pollutants
DIVISION OF MEDICAL SCIENCES
NATIONAL RESEARCH COUNCIL

NATIONAL ACADEMY
OF SCIENCES
WASHINGTON, D.C. 1971

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The members of the study committee were selected for their individual scholarly competence and judgment with due consideration for the balance and breadth of disciplines. Responsibility for all aspects of this report rests with the study committee, to whom sincere appreciation is expressed.

Although the reports of our study committees are not submitted for approval to the Academy membership nor to the Council, each report is reviewed by a second group of scientists according to procedures established and monitored by the Academy's Report Review Committee. Such reviews are intended to determine, *inter alia*, whether the major questions and relevant points of view have been addressed and whether the reported findings, conclusions and recommendations arose from the available data and information. Distribution of the report is permitted only after satisfactory completion of this review process.

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The document has been evaluated *in toto* by every Panel member and represents a team effort under the capable leadership of the Panel Chairman, Dr. Jaroslav J. Vostal. The primary author of the section on natural and industrial sources was Dr. Lawrence Stein; on vegetation, Dr. Leonard H. Weinstein; on animals, Dr. John W. Suttie; on human health, Dr. Frank A. Smith; and on population studies, Dr. Keith Jacobson.

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Preparation of the document has been aided greatly by the availability of various other publications: the 1970 World Health Organization monograph, *Fluorides and Human Health*, and reports and reviews on the effects of airborne fluoride on vegetation by McCune (1969), Brandt (1970), and Weinstein and McCune (1970); on domestic animals by Suttie (1969) and Shupe (1969); and on human health by Smith (1970) and Hodge and Smith (1970). Many chapters were based in part on the above publications or used them as sources of authoritative information.

Charles L. Dunham
Chairman, Division of Medical Sciences

Preface

This document has been prepared at the request of the Air Pollution Control Office (APCO) of the Environmental Protection Agency to provide a scientific basis for APCO to issue an air quality criteria document on airborne fluoride. A concerted effort has been made to evaluate the world literature on the subject and distill the best scientific knowledge available on the biologic effects of fluoride. As the Panel on Fluorides has pursued this goal, it has discovered many areas in which knowledge is incomplete or lacking. It has attempted to evaluate the relative importance of these and in its recommendations for further research has delineated those it considers most important.

The material in any such document is subject to various methods of organization. This panel has chosen to start with a brief introduction, which it is hoped will give the reader familiarity with fluoride and the history of man's understanding of its biologic effects. Following this is an environmental appraisal that treats the sources, both natural and man-made, and then a discussion of the transfer of fluoride in the environment, its physical and chemical changes, its regional and local variations, and its biologic transformations.

To control a pollutant, one must be able to sample and analyze it accurately. This requirement is the subject of the next chapter of the report.

The document next assesses the effects of fluoride on enzymes, cells, and isolated organ and tissue systems and its metabolic effects on intact plants, animals, and humans. Like other pollutants, airborne fluoride can be additive to fluoride accumulated by other routes; therefore, it was considered essential to evaluate the total burden of fluorides in these living systems.

The chapter on population hazards includes a discussion of some population studies, consideration of population groups that may have special susceptibility, and guidelines for occupational and environmental exposure.

Chapter 11 represents the beliefs of the Panel regarding the relative hazards to plants, animals, and humans from airborne fluoride, based on available information. Chapter 12 lists the recommendations for future research.

This document is an attempt to provide a complete treatment of the effects on plants, animals, and humans of airborne fluoride.

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1

Introduction

HISTORY AND PURPOSE

Fluoride is ubiquitous in man's environment, and its beneficial effects in increasing resistance of teeth to cavities have been known since about 1930. Although widespread confidence in the safety and efficacy of artificial fluoridation of water to prevent dental caries is fully justified by the recommendations of the General Assembly of the World Health Organization in 1969, the potential hazard of exposure of large population groups to an increased intake of fluoride has led to continuing controversy among scientists and among the general public.

However, airborne fluoride from dusts and gases has been an obvious health risk in many localities since the time of the earliest known volcanic eruptions. Roholm[549] claims that

> in the Icelandic literature from around about the year 1000 up to recent times we find accounts of how the domestic animals turned sick and died when there were volcanic eruptions. . . . The disease attacked animals which ate the grass contaminated with the fallen ash, which means sheep especially, as cattle and horses were kept stabled as much as possible and so escaped. . . . The symptoms disappeared when the animals were taken indoors and fed on hay mown before the eruption.

But man probably began to affect vegetation and animals when he first baked clay to form brick, and more so when he discovered that some stones heated with ores made the metal flow more readily. In his *De Re Metallica,* Agricola[19] does not mention air pollution, but he does give a good description of the early use of the "fluores" in metallurgy.

Plant damage attributable to industrially produced atmospheric fluoride was recognized by the middle of the nineteenth century,[498] although the source of the first good report on fluoride damage to vegetation is uncertain. Roholm[549] quotes the 1891 report of Mayrhofer[417] on damage to vegetation around a superphosphate factory. Thomas and Alther[671] cite the 1883 report of Schroder and Reuss[576] on damage near a copper smelter; they also credit Reuss[537,671] as the first to distinguish between acute and chronic fluoride injury. In 1907, Ost,[498] in his historical review on the problems covering 60 years or more of work, credits Stöckhart and Schroder[630] with the first detailed investigation of fluoride injury to vegetation and credits the paper of Schroder and Reuss[576] with establishing the necessity of chemical analysis of leaves to confirm injury by fluoride.

By the time Haselhoff *et al.*,[262] Bredemann,[96,97] Roholm,[549] and Romell[554] reviewed the literature, fluoride injury to vegetation as a result of industrial air pollution was well documented. Injury had been reported around superphosphate plants,[417,498,499,540,728] around brickworks,[255,344,624] near glass and enamel factories,[18,498,529] and near a plant manufacturing hydrogen fluoride.[211] By 1920, a new and important source of fluoride injury to vegetation had been recognized near aluminum and other electrochemical reduction plants.[158,200,725]

The effects of the ingestion of fluoride by grazing animals were first clearly summarized by Roholm.[549] The more recent reviews by Davis[167] and Smith[606] concentrate more on experimental studies on laboratory animals.

Thus, evidence on the response of plants to fluoride has been accumulating for more than 100 years, and on the response of animals, for about half that time. The number of industries using or producing fluorine and its compounds increases every year. It is not surprising that estimates of the total atmospheric emissions in 1970 come to nearly 120,000 tons per year in the United States alone. These emissions may be a serious hazard for the future of man and his environment. On the basis of all available evidence and most of the prevailing atmospheric concentrations, fluoride, surprisingly, does not yet constitute a serious problem in pollution of ambient air, so far as toxicity in humans is concerned. Untoward effects in sensitive plant species or grazing animals may, however, occur in limited areas or in the vicinity of fluoride emitters.

The purpose of this document is to summarize and evaluate all available information that describes the extent to which air pollution by fluorine and its compounds can be harmful to plants, animals, and man.

TERMINOLOGY

In the general literature, as well as in chemical publications, the terms "fluorine" and "fluoride" are used interchangeably as generic terms referring to combined forms of the element fluorine. That is true not only in English but also in most foreign literature. Euphony, rather than chemistry, seems to determine the choice.

This document, rather than following common usage, uses the term "fluoride" as a general term everywhere, where exact differentiation between ionic and molecular forms or between gaseous and particulate forms is uncertain or unnecessary. The term covers all combined forms of the element, regardless of chemical form, unless there is a specific reason to stress the existence of a special form of the element–e.g., in references to the gaseous elemental form F_2, in which case the term "fluorine" is used.

Because there can be differences in referring to concentration units given by different investigators, the intention of this document is to designate the exact molecular species and units whenever they can be identified in the original reference. Furthermore, this document reports the work of many investigators in various fields of scientific endeavor, who sometimes use the same or similar terms in different ways. "Parts per million" (ppm) is a common expression of concentration of chemical substance (such as fluoride) in a host substance or suspending medium (soil, plants, food, water, air, etc.). "Parts per billion" (ppb) is also used sometimes; 1 ppm = 1,000 ppb. Concentrations are also expressed as weight of chemical substance (such as fluoride) per unit of weight or volume of suspending medium–for example, milligrams of fluoride per kilogram, or milligrams of fluoride per cubic meter of air; but these terms should not cause the confusion that "parts per million" may.

Concentrations in food, soil, plants, or water, expressed as parts per million, are weights per million weight units of suspending medium. They are readily converted to metric weight units, in that 1 ppm equals 1 mg/kg of soil, food, or plants or 1 mg/liter of water. But "ppm in air" means volumes of contaminant per million volumes of air. The conversion between parts per million in air and milligrams per cubic meter involves some assumptions about a standard atmosphere. The assumptions in the case of studies of the effects of contamination on plant life are different from those in the case of studies of health effects on experimental animals or industrial workers.

In reporting air concentrations affecting vegetation, the following assumptions of the standard atmosphere are made: pressure, 760 torr; temperature, 288 K (15 C); and volume of a mole of a gas, 23.645 liters. In reporting air concentrations affecting experimental animals and in industrial-hygiene surveys, the standard atmosphere is considered to have the same pressure, but a temperature of 298 K (25 C) and thus a volume of a mole of gas of 24.45 liters.

In the study of effects of airborne fluoride on plants, parts per million and milligrams per cubic meter are related by the following formula:

$$\text{concentration in ppm} = \text{concentration in mg/m}^3 \times \frac{23.645}{\text{mol.wt.}};$$

and in the study of effects on experimental animals and man by:

$$\text{concentration in ppm} = \text{concentration in mg/m}^3 \times \frac{24.45}{\text{mol.wt.}}.$$

The reader should also be alert to the molecular species intended by the investigator. Some investigators treat fluorine as F, with a molecular weight of 19; others treat fluorine as F_2, with a molecular weight of 38. Some investigators treat fluoride as F; others, as F in terms of HF. Wherever possible and appropriate in this review, concentrations in parts per million or parts per billion will also be expressed in milligrams per cubic meter or micrograms per cubic meter, and the molecular species will be identified.

2

Environmental Sources and Forms of Fluoride

NATURAL SOURCES OF FLUORIDE IN THE ENVIRONMENT

Minerals and Soils

Fluorine chemically bound as fluoride is widely distributed in the earth's crust. It occurs in both igneous and sedimentary rocks and is estimated to constitute between 0.06% and 0.09% by weight of the upper layers of the lithosphere.[342,360] The chief fluorine-containing minerals are fluorspar (CaF_2), cryolite (Na_3AlF_6), and fluorapatite [$Ca_{10}F_2(PO_4)_6$]. For many years, cryolite was obtained from a large natural deposit in Greenland, at Ivigtut, on the south side of Arsuk fjord. However, this source has been largely depleted by mining, and synthetic cryolite for industrial purposes is now manufactured in chemical plants. Several small deposits of cryolite are known (in Colorado, in the Ural Mountains, and in the Pyrenees), but the mineral cannot be extracted economically, and the deposits are therefore of little commercial importance. Fluorspar and fluorapatite are widespread and are mined in the United States and many other countries. A 500-square-mile area at Bartow, Florida, contains very extensive deposits of phosphate rock consisting chiefly of fluorapatite and is one of the largest centers for the manufacture of phosphate fertilizers.

Soils may contain fluoride in the form of several different minerals, including biotite, muscovite, hornblende, apatite, and fluorite (fluorspar). Analytic profiles of 30 soils collected from different parts of the United States indicate that the fluoride content usually increases with depth.[548] The fluoride content was found to range from 20 to 500 ppm in samples taken from depths of 0–3 in. (average, 190 ppm) and from 20 to 1,620 ppm in samples taken from depths of 0–12 in. (average, 292 ppm). Some soils have been found to contain unusually high concentration of fluoride, such as an Idaho soil with 3,870 ppm[548] and a Tennessee soil with 8,300 ppm[388] of fluoride.

Natural Waters

The fluoride content of natural waters varies widely in different areas and depends on such factors as the source of the water (surface or subterranean), type of geologic formation present, amount of rainfall, and amount of water lost by evaporation. Surface waters obtained from rivers and lakes generally contain lower concentrations of fluoride than spring and well waters. Many states along the northeastern seaboard of the United States have water supplies with natural concentrations of fluoride ranging from only 0.02 to 0.1 ppm (average values).[139] In western, midwestern, and southern states, the average concentrations are more often above 0.2 ppm.[62,206] The highest concentrations are observed in areas where the soil is rich in apatite or other fluoride minerals and the water is obtained from wells. In these areas, concentrations of 1–7 ppm are not uncommon. Mottled tooth enamel is frequently observed in the population when the fluoride concentration of the water is very high, such as 2–7 ppm or more. The incidence of mottled enamel drops sharply, however, when the fluoride concentration is reduced to 1 ppm or less, either by the use of an alternative water supply or by chemical removal of excess fluoride.[184,236] Natural waters containing high concentrations of fluoride are found in many parts of the world, including Europe, Asia, Africa, North America, and South America.[139] The effects on the population have been studied in India, Japan, Italy, France, England, Germany, the Soviet Union, the United States, and other countries. Exceptionally high fluoride concentrations (53 and 93 ppm) have been reported from some regions of South Africa and

North Africa.[139] In comparison with that from many inland sources, seawater contains very little fluoride, approximately 1.4 ppm.

Volcanoes and Fumaroles

Appreciable amounts of gaseous and solid fluorides are discharged into the atmosphere by active volcanoes and fumaroles. The emissions have been studied chiefly in Italy, Japan, New Zealand, Mexico, the Soviet Union, and the United States, but the effects have also been noted in many other regions where there are active volcanoes. In the Valley of Ten Thousand Smokes (Katmai region of Alaska), hydrogen fluoride has been identified as one of the principal fluoride emissions, and in fumes from Vesuvius and Vulcano in Italy, many products have been identified, including hydrogen fluoride, ammonium fluoride (NH_4F), silicon tetrafluoride (SiF_4), ammonium fluorosilicate [$(NH_4)_2SiF_6$], sodium fluorosilicate (Na_2SiF_6), potassium fluorosilicate (K_2SiF_6), and potassium fluoroborate (KBF_4). Fluorides have also been reported in gaseous and solid emissions from the following volcanoes: Asama and Mihara in Japan,[488] Oyama on Miyake Island,[489] Satsuma on Iwo Jima,[734,734] and Klyuchevskoi in the Soviet Union.[337] The amounts of volatile fluorine, chlorine, and sulfur compounds emitted by Asama increased greatly in the weeks preceding two eruptions in 1957. The studies of Mt. Asama, which have been carried on since 1933, suggest that eruptions of active volcanoes can be predicted by measuring the local concentrations of gaseous fluorine compounds and other compounds in air.[488]

Foods

Virtually all foods contain trace amounts of fluoride, but the total daily intake in any average human diet is low. In the United States, the daily intake of fluoride by an adult in an average general diet is estimated to be 0.2–0.3 mg.[139] To this must be added the fluoride ingested in drinking water, which may be 1.0 mg or more. Most plants absorb very little fluoride from the soil[388,671] even when the fluoride content of the soil is naturally high or is artificially increased by the addition of chemicals, such as barium fluorosilicate ($BaSiF_6$), sodium fluorosilicate, calcium fluoride (CaF_2), magnesium fluoride (MgF_2), potassium fluoride (KF), sodium fluoride (NaF), or hydrofluoric acid (HF). (Tea is a notable exception, in that it takes up fluoride from soil and may contain as much as 400 ppm on a dry-weight basis.) However, soluble fluoride is often localized in the leaves. Most plants contain between 0.1 and 10 ppm of fluoride on a dry-weight basis. The following ranges of fluoride concentration have been reported[139] in some typical fresh foods in parts per million: meats, 0.01–7.7; fish, less than 0.10 to 24; citrus fruits, 0.04–0.36; noncitrus fruits, 0.02–1.32; cereals and cereal products, less than 0.10 to 20; vegetables and tubers, 0.10–3.0; milk, 0.04–0.55; beer, 0.15–0.86; wine, 0.0–6.34; eggs, 0.00–2.05; butter, 0.4–1.50; cheese, 0.13–1.62; sugar, 0.10–0.32; and coffee, 0.2–1.6.

A number of plant species in Australia, South America, and Africa have been shown to synthesize fluoroaliphatic acids under natural conditions (see Chapter 3). The toxic effects of these species are due to organic acids and not to fluoride ions.[512]

INDUSTRIAL SOURCES OF FLUORIDE IN THE ENVIRONMENT

Fluoride dusts and gases are emitted to the atmosphere by many types of industrial operations. These include the grinding, drying, and calcining of fluoride-containing minerals, acidulation of the minerals, smelting and electrochemical reduction of metals with fluoride fluxes or melts, kiln-firing of brick and ceramic materials, melting of raw materials in glassmaking, and use of fluoride-containing chemicals for cleaning, etching, and electroplating in manufacturing processes. The combustion of coal also introduces fluoride pollutants into the atmosphere. Some of the chief industrial processes that release fluoride dusts and gases to the atmosphere are described in this section. The magnitudes of the fluoride emissions are listed in Table 2-1, if known. Water fluoridation is briefly discussed, and some fluorine-containing drugs, anesthetics, and pesticides are described.

Manufacture of Phosphate Fertilizers and Wet-Process Phosphoric Acid

Most of the phosphate rock mined in the United States is converted into phosphate fertilizers (normal superphosphate, triple superphos-

TABLE 2-1 Estimated Total Fluoride Emissions from Major Industrial Sources in the United States in 1968[a]

Source	Atmospheric Emissions, tons/year
Manufacture of normal superphosphate fertilizer	9,700
Manufacture of wet-process phosphoric acid	3,000
Manufacture of triple superphosphate fertilizer	300
Manufacture of diammonium phosphate fertilizer	100
Manufacture of elemental phosphorus	5,500
Manufacture of phosphate animal feed	100
Manufacture of aluminum	16,000
Manufacture of steel (open-hearth furnace)	16,800
Manufacture of steel (basic-oxygen furnace)	8,400
Manufacture of steel (electric furnace)	14,900
Welding operations	2,700
Nonferrous-metal foundries	4,000
Manufacture of brick and tile products	18,500
Manufacture of glass and frit	2,700
Combustion of coal	16,000
Total	118,700

[a] Data from U.S. Department of Health, Education, and Welfare.[686]

phate, and ammonium phosphate types), phosphoric acid, and elemental phosphorus. The elemental phosphorus is then used to manufacture a variety of phosphorus compounds. During grinding and drying of the phosphate rock, which consists chiefly of fluorapatite and contains 3–4% fluorine, considerable amounts of dust are produced. Recovery and control devices—such as dry cyclones, filters, electrostatic precipitators, and wet scrubbers—are used to limit the dust emissions to the atmosphere. The efficiency of the various control systems varies from about 95% to 99.9%.[687]

Silicon tetrafluoride is the chief gaseous pollutant released to the atmosphere in many of the manufacturing steps; phosphate rock contains silica (SiO_2), as well as fluorapatite. In the manufacture of normal superphosphate fertilizer, ground phosphate rock is treated with sulfuric acid,[602] and both hydrofluoric acid and silicon tetrafluoride are formed. The reactions can be written as follows:

$$Ca_{10}F_2(PO_4)_6 + 7H_2SO_4 + 3H_2O \rightarrow 3CaH_4(PO_4)_2 \cdot H_2O + 7CaSO_4 + 2HF\uparrow .$$

and

$$SiO_2 + 4HF \rightarrow SiF_4 + 2H_2O.$$

Hydrofluoric acid and silicon tetrafluoride are also formed in the production of wet-process phosphoric acid and triple superphosphate fertilizer (from the phosphoric acid). The two initial reactions are as follows:

$$Ca_{10}F_2(PO_4)_6 + 10H_2SO_4 + 20H_2O \rightarrow 6H_3PO_4 + 10CaSO_4 \cdot 2H_2O + 2HF\uparrow,$$

and

$$Ca_{10}F_2(PO_4)_6 + 14H_3PO_4 + 10H_2O \rightarrow 10CaH_4(PO_4)_2 \cdot H_2O + 2HF\uparrow.$$

Again, volatile silicon tetrafluoride is released through the reaction of hydrofluoric acid with silica in the phosphate rock.

In modern plants that practice good emission control, the off-gases are passed through water-spray towers or other types of scrubbers. Most of the silicon tetrafluoride is removed through reaction with water to form aqueous fluorosilicic acid (H_2SiF_6). Hydrogen fluoride vapor is also removed by the scrubbing system through formation of aqueous hydrofluoric acid. However, the efficiency of emission control varies widely from one plant to another, and some of the older plants permit the escape of large amounts of silicon tetrafluoride and hydrogen fluoride to the atmosphere.

In the manufacture of wet-process phosphoric acid, waste liquors and slurries are produced that contain gypsum ($CaSO_4 \cdot 2H_2O$), sodium and potassium fluorosilicates, hydrofluoric acid, and fluorosilicic acid. In coastal areas, these are often disposed of in the sea. Large settling ponds are constructed in other areas, and the wastes are allowed to accumulate in the ponds. The concentrations of hydrofluoric and fluorosilicic acids in the ponds gradually increase when the waters are recirculated into the manufacturing plants for reuse.

Volatile fluorides vaporizing from the ponds are not controlled and may constitute 90% or more of the total fluoride emissions to the atmosphere.[686] Water runoff from the ponds through accidental leakage or overflow in times of heavy rain is also a problem, because it can spread the acid solutions into adjacent areas. The acids are usually neutralized and the fluorides precipitated by additions of lime or limestone before runoff into streams is permitted.

Diammonium phosphate ($2NH_3 \cdot H_3PO_4$) fertilizer is one of the major types now produced in the United States; it is a granular powder, high in plant nutrients, with low shipping cost based on phosphorus pentoxide (P_2O_5) content. In the production process, phosphoric acid solution is progressively neutralized with ammonia, and water is removed by evaporation. Fluoride emissions are lower than those in normal superphosphate, triple superphosphate, and wet-process phosphoric acid production, because the initial reaction involves phosphoric acid, rather than fluorapatite.

Plants manufacturing normal superphosphate fertilizer are scattered throughout the United States, although most are in the central and southeastern states.[686] Triple superphosphate fertilizer plants have a more limited distribution; the principal ones are in Florida, Mississippi, North Carolina, Missouri, Idaho, Utah, and California. Wet-process phosphoric acid plants and diammonium phosphate plants are widespread, with most near phosphate mines (in Florida, North Carolina, Idaho, and California) or waterways where phosphate rock can be shipped in cheaply by barge (in Texas, Louisiana, Mississippi, Illinois, and Missouri).

The following estimates have been made of the amounts of fluoride released to the atmosphere in 1968 in the manufacture of each of these products:[686] normal superphosphate fertilizer, 9,700 tons; triple superphosphate fertilizer, 300 tons; wet-process phosphoric acid, 3,000 tons; and diammonium phosphate fertilizer, 100 tons.

Manufacture of Phosphorus and Phosphate Feed Supplements

Elemental phosphorus is produced in the United States in amounts exceeding 600,000 tons annually by an electric-furnace process. Ground phosphate rock is first sintered or passed through a heated rotary kiln to produce an agglomerate of appropriate size, then mixed with silica and coke, and finally heated in a carbon-lined furnace with carbon electrodes. The following reaction occurs:[691]

$$2Ca_{10}F_2(PO_4)_6 + 18SiO_2 + 30C \rightarrow 18 \cdot CaO \cdot SiO_2 \cdot \frac{1}{9} CaF_2 + 30CO + 3P_4 \uparrow.$$

Carbon monoxide and phosphorus are evolved as gases, together with silicon tetrafluoride formed through a secondary reaction of silica and fluorapatite. Some particulate matter is also released. The phosphorus is condensed in a water-spray tower, and the carbon monoxide is either used as fuel or burned as a waste product. The water condensate contains hydrated silica, fluorosilicates, and other impurities, as well as the desired phosphorus.

Escape of silicon tetrafluoride to the atmosphere can occur by leakage of gases from the furnace through seals around the electrodes or through the supply chutes for feed materials. The leaks are largely eliminated in modern furnace design. Silicon tetrafluoride, hydrogen fluoride (HF), and dusts are evolved during the initial heat-treatment step, in which the agglomerate is formed. These are largely removed by spray towers, wet-cyclone scrubbers, or venturi scrubbers in modern plants, but older plants still emit substantial amounts to the atmosphere. The slag that is periodically tapped from the furnace as a liquid contains about 3% fluoride, as well as many metal oxides. This is either solidified and discarded as a waste product or granulated with a water jet and sold for use as an agricultural liming agent or for other purposes.

The total amount of fluoride emitted to the atmosphere in elemental phosphorus production in the United States has been estimated at 5,500 tons in 1968.[686]

In the preparation of animal feed supplements, phosphate rock is processed to remove most of its fluorine content. The phosphate rock is treated with phosphoric acid and in some instances with other additives, such as soda ash, and is then heated in a drying kiln at temperatures of 1100–1350 C. The fluorine is evolved chiefly as hydrogen fluoride: silicon tetrafluoride is converted to hydrogen fluoride and nonvolatile silica by reaction with water vapor at these high temperatures. At least 95% of the fluorine must be removed for the final granulated product to be suitable for animal feed, because the concentration of residual fluorine determines the amount of the supplement that can be safely added to an animal diet without producing symptoms of fluorosis. Various types of scrubbers are used to collect the hydrogen fluoride and dusts from the drying kiln.

The total emission of fluoride to the atmosphere from the manufacture of phosphate feed supplements in the United States has been estimated at 100 tons in 1968.[686]

Manufacture of Aluminum

Aluminum is now produced almost entirely by the Hall-Heroult process, in which alumina (Al_2O_3) is dissolved in molten cryolite and reduced electrolytically. The alumina is obtained from bauxite ore, which is found in many parts of the world, including Europe, Africa, North and South America, and Australia. Originally, cryolite was obtained only from natural deposits in Greenland, but this supply proved inadequate for world needs, and it is now synthesized. In addition to cryolite, the melt usually contains calcium fluoride, which lowers the melting point to below 1000 C.

The electrolytic cell contains a carbon lining, which serves as both the cathode and the container for the melt. A layer of firebrick insulation surrounds the carbon lining. In one type of cell (a prebake cell), carbon anode blocks are suspended in the melt and individually replaced as they are consumed. In another type (known as a Soderberg cell), the anode is formed by a paste made of carbon and pitch, which is fed in continuously from an external source and baked to a solid before it enters the cell. Aluminum is formed by reduction at the cathode and floats as a molten layer above the electrolyte. The aluminum in turn is covered by a surface layer of solid alumina. Oxygen liberated at the anode by electrolysis reacts with the anode and forms a mixture of carbon monoxide and carbon dioxide, which escapes from the cell.

The net chemical change is approximately that shown in the following equation:

$$Al_2O_3 + 2C \rightarrow 2Al + CO\uparrow + CO_2\uparrow.$$

In addition to carbon monoxide and carbon dioxide, gaseous emissions from aluminum electrolysis cells include sulfur dioxide (SO_2), silicon tetrafluoride, hydrogen fluoride, carbonyl sulfide (COS), carbon disulfide (CS_2), hydrocarbons, and water vapor.[686] Particulate emissions include alumina, carbon, cryolite, aluminum fluoride (AlF_3), calcium fluoride, chiolite ($Na_5Al_3F_{14}$), and iron oxide (Fe_2O_3). In the past, especially during World War II, when aluminum was in great demand for the production of aircraft, emissions were not controlled and were released to the atmosphere. The harmful dusts and gases from aluminum plants are now largely removed by filters, centrifugal collectors, electrostatic precipitators, and various types of wet-scrubbing systems.[668]

The total amount of fluoride emissions to the atmosphere by aluminum manufacturing plants in the United States has been estimated at approximately 16,000 tons in 1968.[686]

Manufacture of Steel

Fluorspar is used as a flux in open-hearth, basic-oxygen, and electric-furnace steelmaking operations to increase the fluidity of the slags and enhance the removal of phosphorus and sulfur impurities from the melts.[173,437] Phosphorus is oxidized to the pentoxide, which combines with lime (CaO) in the slag; if sufficient fluorspar is present, the phosphorus may also form fluorapatite in the slag.

In electric-furnace steelmaking, two refining stages are generally used. An oxidizing slag is formed in the initial step and then removed from the surface of the molten metal and replaced by a reducing slag containing lime, silica, fluorspar, and coke (C). The second slag allows sulfur impurity to be mostly removed and lowers the oxygen content of the melt. For collection of fumes, one type of electric furnace is made with an overhead duct ("snorkel") in the roof, through which the fumes are carried to scrubbing devices. Others have external hoods mounted above the furnaces for the same purpose.

In the basic-oxygen process, a jet of pure oxygen is blown into the vessel containing the melt through a water-cooled "lance," which is placed just above the surface of the melt. Dense fumes are evolved during the blowing period and are carried through ducts to cleaning devices, such as electrostatic precipitators, cloth filters, and venturi scrubbers.

The efficiency of dust and gas removal in the individual devices ranges from approximately 95% to 99%. Fluorides constitute only a small fraction of all the fumes produced and are believed to consist chiefly of calcium fluoride and hydrogen fluoride. A fairly complete description has been given of the measures used in one steel plant to reduce fluoride emissions from both a sintering operation (for pretreatment of a fluoride-containing ore) and an open-hearth process.[232]

The following estimates have been made of the total amounts of fluoride emitted to the atmosphere in the United States by each of the three steelmaking processes in 1968:[686] open-hearth, 16,800 tons; basic-oxygen, 8,400 tons; and electric-furnace, 14,900 tons.

Manufacture of Brick, Tile, Pottery, and Cement

Clays used in the manufacture of brick, tile, pottery, and cement generally contain 0.02–0.3% of fluorine in the form of hydrated micas, such as muscovite and illite. For production of brick, tile, and pottery ware, clays of suitable composition are shaped in either wet or dry form and fired in kilns at gradually increasing temperatures (up to approximately 1100 C). During the firing process, gaseous hydrogen fluoride and silicon tetrafluoride are liberated. Estimates of the amounts released in brickmaking range from approximately 30% to 95% of the fluorine initially present in the clays.[80,580] Gases and dusts from the kilns are usually discharged directly to the atmosphere without any cleanup. Many instances are known of injuries to plants and animals caused by the emissions, particularly in Europe.

The total amount of fluoride emission from brick, tile, and pottery manufacture in the United States in 1968 has been estimated at 18,500 tons.[686]

In the manufacture of Portland cement, mixtures of clay and limestone are gradually heated to about 1450 C in rotary kilns to form clinker, which is then ground to fine powder with a small amount of gypsum. The gypsum controls the setting time of the cement. Fluorine in the clay reacts with lime during the firing, forming calcium fluoride. This is chiefly incorporated in the clinker and the final product. The operations produce large amounts of dust, which in modern plants is collected by cyclones and glass-fabric filters. If the dust collection is efficient, fluoride emission to the atmosphere is very low. In the manufacture of "white cements," cryolite and fluorspar are often added as fluxing agents; dust from the "white cements" therefore contains a higher percentage of fluoride than dust from ordinary Portland cement.

Manufacture of Glass, Enamel, and Fiber Glass

Silicate glasses are produced by combining and melting raw materials that contain silica, metal carbonates, or metal oxides. One of the most widely used types, soda-lime glass, has a composition of approximately 72% silica, 15% soda (Na_2O), 10% lime and magnesia (MgO), 2% alumina, and 1% miscellaneous other oxides.[301] Borosilicate glass, which has a low coefficient of thermal expansion and is used extensively for scientific apparatus and baking ware, contains boric oxide (B_2O_3) in place of most of the soda and little or no magnesia or lime. Fluorine-containing minerals and compounds–such as fluorspar, cryolite, and sodium fluorosilicate–are frequently added to glasses that are to be used for special purposes. At the high temperatures used to melt the raw materials (1450–1600 C in most instances), carbon dioxide is evolved. Gaseous and particulate fluorides are also evolved if fluoride chemicals are present. Fabric filters are chiefly used to remove particulate matter from the off-gases.

Solutions of hydrofluoric acid or ammonium bifluoride ($NH_4F \cdot HF$) are often used to clean or etch the products made in glassworks. The frosted inner surface of light bulbs, for example, is produced by an etching process. Vapors from the etching solutions are further sources of fluoride emissions to the atmosphere if control procedures are absent or inadequate.

For the production of enamel coatings on metal, commonly known as porcelain enamel or vitreous enamel, special types of glass are made. The glass may include calcium fluoride, aluminum fluoride, sodium fluoride, and sodium or potassium fluorosilicate, as well as silica, boric oxide, soda, potash (K_2O), alumina, and other metal oxides. The molten glass is quenched with air or water, causing it to solidify and fracture into small particles called frit. The frit is ground to powder in a ball mill, mixed with additives, and applied to a suitably prepared metal surface, such as steel or cast iron, in either wet or dry form (slip, slurry, or powder coating). The coated metal is then fired in a furnace at 800–950 C to fuse the enamel and bond it to the surface.

During the melting of the various types of glass used in making glassware and frit, approximately 20% of the fluorine introduced in the raw materials may be lost as gaseous fluorine compounds and dusts,[720] such as silicon tetrafluoride, hydrogen fluoride, calcium fluoride, aluminum fluoride, sodium fluoride, and sodium fluorosilicate. The losses during the firing of enamels are highly variable, depending on batch compositions and firing temperatures. Fabric filters or water venturi scrubbers are usually used to control emissions from frit smelters, but the efficiency of fluoride removal is often less than 90%.

From the consumption of fluorspar in glass and frit manufacture, the total fluoride emission to the atmosphere from this industry in the United States in 1968 has been estimated at 2,700 tons.[686]

Silicate materials are also converted to fiber glass mats for thermal insulation and fibers for textile manufacture on a large industrial scale. The manufacturing processes involve blowing fine streams of molten glass with air or steam into fibers, which are collected in mats or wound as continuous filaments. Fluoride emissions are produced in the furnaces, during melting of the glass charges, and in the blow-chambers. Chiefly particulate emissions are controlled in this industry by the use of fabric filters.

Combustion of Coal

Samples of coal from various parts of the world have been analyzed and found to contain 0.001–0.048% fluoride, usually as fluorapatite or fluorspar.[2] An average fluoride content of 0.008% has been found in samples from 83 mines in the United States.[1] During combustion, approximately half the fluoride in the coal is evolved as gaseous hydrogen fluoride, silicon tetrafluoride, and particulate matter. From the total of 402.3 million tons of coal burned in the United States in 1968 for steam and energy production, fluoride emissions to the atmosphere have been estimated at 16,000 tons.[686] An additional 96.5 million tons of coal was used in the United States in 1968 for coke and coal-gas manufacture, but emissions from this source were considered to be negligible.

Most of the emissions occur in coal-burning electric-power plants. At present, electrostatic precipitators are generally used to remove particulate matter from the combustion products, but gases are released to the atmosphere. Sulfur oxides constitute a more serious pollution problem from the plants than fluorides, inasmuch as the amounts of sulfur are approximately 100 times greater than the amounts of fluoride in coal.

Manufacture of Fluorides

Hydrogen fluoride is manufactured on a major industrial scale by heating fluorspar with sulfuric acid. The hydrogen fluoride is liberated as a vapor at temperatures between 100 and 250 C and is condensed, purified, and stored as a very volatile liquid in steel tanks and cylinders. The decomposition of the fluorspar takes place as follows:

$$CaF_2 + H_2SO_4 \rightarrow CaSO_4 + 2HF\uparrow .$$

In addition to hydrogen fluoride, such gases as sulfur dioxide, silicon tetrafluoride, and carbon dioxide are produced by decomposition of impurities in the fluorspar.[216] The off-gases from the condensers are passed through successive scrubbers containing sulfuric acid and water, which recover most of the silicon tetrafluoride as aqueous fluorosilicic acid. Carbon dioxide and sulfur dioxide pass through the scrubbers and are generally vented to the atmosphere. In 1968, 284,000 tons of hydrogen fluoride were produced in the United States for various purposes. Most industries use hydrogen fluoride as the anhydrous liquid or gas. A small fraction of the total production is converted to aqueous hydrofluoric acid containing 48%, 60%, or 70% hydrogen fluoride. The anhydrous liquid is frequently shipped to industrial users in 6,000- and 11,000-gal tank cars; because its vapor pressure is greater than 1 atm at 25 C, great care must be used in loading and unloading the bulk quantities to prevent escape of the vapor to the atmosphere.

Elemental fluorine is produced by electrolysis of solutions of potassium bifluoride (KHF_2) in anhydrous hydrogen fluoride. Fluorine gas (F_2) is liberated at the anode, and hydrogen gas (H_2) at the cathode. Because the anode is made of carbon, some fluorocarbon impurities are produced, although the major impurity in the fluorine is hydrogen fluoride. Most of the latter is removed by passing the fluorine through absorption towers packed with sodium fluoride, which reacts with the hydrogen fluoride to form sodium bifluoride ($NaHF_2$). The absorption towers are periodically purged in the reverse direction with heated air to regenerate the sodium fluoride, and the small amounts of fluorine evolved with hydrogen fluoride in this step are burned with propane gas. The off-gases then pass through scrubbers containing sodium carbonate (Na_2CO_3) for removal of hydrogen fluoride before the residual gases are vented to the atmosphere.

In recent years, cryogenic trailers have been developed for shipment of bulk amounts of liquid fluorine by truck. One type of trailer now in use has a capacity of 2.5 tons of liquid fluorine. The fluorine is contained in an inner vessel surrounded by liquid nitrogen and an outer insulating jacket.[475] Because fluorine is one of the most reactive elements known and is highly toxic, great care is required in loading, transporting, and unloading the liquid cargoes.

From hydrogen fluoride and fluorine, chemical companies now manufacture fluorides of almost all metal and nonmetal elements for commercial and experimental use. The manufacture of fluorocarbon compounds, such as Freon propellants and fluorinated plastics, has grown into a particularly large chemical industry. In 1968, approximately 40% of all the hydrogen fluoride manufactured in the United States was converted, by various steps, into fluorocarbon compounds and products.[686] The compounds include dichlorodifluoromethane (CCl_2F_2), trichlorofluoromethane (CCl_3F), tetrafluoromethane (CF_4), tetrafluoroethylene (C_2F_4), vinyl fluoride (C_2H_3F), and hexafluoropropylene (C_3F_6), and the products include many polymers derived from these and similar compounds. Fluorocarbon compounds generally have low toxicity, high stability, and other properties that make them suitable for use as aerosol propellants and working fluids in refrigeration equipment. However, some precautions are necessary when the compounds are used in confined spaces, because inhalation of high concentrations of the vapors is hazardous to health.[182] The methods of manufacture include electrolysis of organic compounds in hydrogen fluoride solutions;[471] reaction of hydrocarbons with metal fluorides, such as cobalt trifluoride (CoF_3) and silver difluoride (AgF_2);[263] reaction of halocarbons containing chlorine, bromine, or iodine with hydrogen fluoride in the presence of catalysts; and pyrolysis of unsaturated fluorocarbons.

A number of halofluorocarbons and fluorinated ethers have been tested in animal experiments for their effectiveness as anesthetics.[345,355] Many of those tested proved to be moderately or highly toxic. Among those shown to be safe for use with humans and now in general clinical use as anesthetics are 1,1,1-trifluoro-2,2-bromochloroethane ($CF_3CHBrCl$), 2,2,2-trifluoroethyl vinyl ether ($CF_3CH_2{-}O{-}CH{=}CH_2$), and 2,2-dichloro-1,1-difluoroethyl methyl ether ($CH_3{-}O{-}CF_2CHCl_2$).

Several commercial processes have been developed for the manufacture of synthetic cryolite. In one process, a solution of sodium aluminate ($NaAlO_2$) and sodium hydroxide (NaOH) is treated with anhydrous hydrogen fluoride or aqueous hydrofluoric acid to form a water suspension of cryolite:[214]

$$NaAlO_2 + 2NaOH + 6HF \rightarrow Na_3AlF_6 + 4H_2O.$$

The suspension is filtered or centrifuged, and the cryolite cake is dried and calcined at 500–700 C. Sodium carbonate may be substituted for the sodium hydroxide, and aluminum sulfate [$Al_2(SO_4)_3$] low in iron impurity may also be substituted for the sodium aluminate:

$$NaAlO_2 + Na_2CO_3 + 6HF \rightarrow Na_3AlF_6 + CO_2 + 3H_2O;$$

$$Al_2(SO_4)_3 + 6Na_2CO_3 + 12HF \rightarrow 2Na_3AlF_6 + 3Na_2SO_4 + 6CO_2 + 6H_2O.$$

In a second process, cryolite is obtained as a valuable by-product from the fluorosilicic acid wastes of phosphorus and phosphate fertilizer plants.[659] Silica is first precipitated from the acid solutions by addition of ammonia (NH_3). The aqueous ammonium fluoride that is formed is then added to a solution of sodium sulfate (Na_2SO_4) and aluminum sulfate to produce cryolite:

$$12NH_4F + 3Na_2SO_4 + Al_2(SO_4)_3 \rightarrow 2Na_3AlF_6 + 6(NH_4)_2SO_4.$$

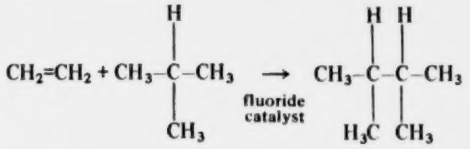
Large amounts of cryolite are also recovered from the carbon linings of electrolysis cells used in the manufacture of aluminum. When the cells are shut down for maintenance, the linings frequently contain 10–30% of cryolite by weight. The linings are crushed and treated with sodium hydroxide or sodium carbonate solution to extract the aluminum and fluoride values, and the cryolite is then reprecipitated by the addition of carbon dioxide. The steps are as follows:[214]

$$Na_3AlF_6 + 4NaOH \rightarrow 6NaF + NaAlO_2 + 2H_2O;$$

$$6NaF + NaAlO_2 + 2CO_2 \rightarrow Na_3AlF_6 + 2Na_2CO_3.$$

Petroleum Refining

Hydrogen fluoride is widely used by the petroleum industry as an alkylation catalyst for producing high-octane gasolines. Boron trifluoride (BF_3) is also used for this purpose, and hydrogen fluoride and boron trifluoride are sometimes used together.[49] In an alkylation reaction, a paraffin hydrocarbon or an aromatic compound is added to the double bond of an olefin to form a compound of higher molecular weight. The reaction of ethylene ($CH_2{=}CH_2$) with isobutane [$(CH_3)_3CH$] to form 2,3-dimethylbutane [$(CH_3)_2CHCH(CH_3)_2$] is a typical example:



$$CH_2{=}CH_2 + CH_3{-}\overset{\overset{\displaystyle H}{|}}{\underset{\underset{\displaystyle CH_3}{|}}{C}}{-}CH_3 \xrightarrow[\text{catalyst}]{\text{fluoride}} CH_3{-}\overset{\overset{\displaystyle H}{|}}{\underset{\underset{\displaystyle H_3C}{|}}{C}}{-}\overset{\overset{\displaystyle H}{|}}{\underset{\underset{\displaystyle CH_3}{|}}{C}}{-}CH_3$$

By reactions of this type, low-boiling isoparaffins are converted to high-boiling isoparaffins with improved antiknock properties. The reactions are carried out in large pressure vessels, with reactants and catalysts in either the gas or liquid state. Volatile fluorides may be released to the atmosphere during loading and unloading operations (for example, during purging of transfer lines from hydrogen fluoride tank cars) and during disposal of waste products. Tarry residues that liberate fluoride fumes on exposure to air are frequently produced.

Metal Casting, Welding, and Brazing

In magnesium and aluminum foundries, fluorides are used as fluxes and as coatings for sand molds in metal-casting operations. Among the compounds used for this purpose are ammonium fluoride, ammonium bifluoride, ammonium fluoroborate (NH_4BF_4), potassium fluoroborate, and ammonium fluorosilicate. On contact with the hot metals, the compounds vaporize and partially decompose into such products as hydrogen fluoride, silicon tetrafluoride, and boron trifluoride. Although fumes of the compounds and decomposition products are removed from the work areas by ventilating fans, efforts are seldom made to reduce fluoride emissions to the atmosphere by wet scrubbers or other devices.

In arc welding of ferrous and nonferrous metals, it is now common practice to use electrodes coated with calcium fluoride or other inorganic fluorides, which serve as fluxing agents at the high temperatures generated by the arc. Gaseous compounds, such as hydrogen fluoride and silicon tetrafluoride, and particulate fluorides are released when the coated electrodes are used.[258] In a recent study of fluoride emissions from welding in shipyards and large boiler plants, approximately 300 air samples from welding sites were analyzed.[612] The fluoride concentrations exceeded 2.5 mg/m^3 (the recommended threshold limit value for an 8-hr day) in 37% of samples from enclosed areas and 13% of samples from semienclosed and open areas. During tests conducted in a simulated "average welding shop" with electrodes from five manufacturers,[324] concentrations of particulate fluoride ranging from 0.03 to 1.5 mg/m^3 were found in air samples from the inside of welders' head shields and concentrations ranging from 10.9 to 19.0 mg/m^3 were found in samples from sites outside the shields (areas of dense fumes). Fumes from industrial welding operations of this type usually are removed only by ventilation and vented directly to the atmosphere.

Fluxes containing metal fluorides are also used in silver soldering and other types of metal brazing. Brazing is performed in many manufacturing operations and is a further source of gaseous and particulate fluoride pollutants in the atmosphere in some industrial districts.

Operation of Atomic Energy Installations and Production of Uranium Hexafluoride

Gaseous diffusion plants that use uranium hexafluoride (UF_6) to separate uranium isotopes have been built in the United States, England, the Soviet Union, and France. The plants produce ^{235}U, which undergoes fission with slow neutrons, for nuclear weapons and also produce uranium enriched in ^{235}U for nuclear power reactors of various types. An elaborate technology has been developed in each of these countries for the production and handling of uranium hexafluoride and several lower fluorides, such as UF_3, UF_4, U_4F_{17}, U_2F_9, and UF_5.

Uranium hexafluoride is usually produced from uranium oxides in several steps, with uranium tetrafluoride (UF_4) as an intermediate; inexpensive hydrogen fluoride can be used to prepare the tetrafluoride. In one method, uranium trioxide (UO_3) is reduced to the dioxide (UO_2) with hydrogen at approximately 700 C; the dioxide is then converted to the tetrafluoride with hydrogen fluoride at 500 C, and uranium hexafluoride is formed through the reaction of the tetrafluoride with fluorine at 350 C:[95,291,325,578,604]

$$UO_3 + H_2 \xrightarrow[700\ C]{} UO_2 + H_2O,$$

$$UO_2 + 4HF \xrightarrow[500\ C]{} UF_4 + 2H_2O,$$

and

$$UF_4 + F_2 \xrightarrow[350\ C]{} UF_6.$$

Only one third of the total fluorine in uranium hexafluoride is provided by relatively expensive elemental fluorine in this method.

Several other methods for producing uranium hexafluoride with halogen fluoride reagents (ClF_3, BrF_3, and BrF_5) have also been developed.[69,303,320,627] The halogen fluorides are capable of converting uranium oxides or uranium metal directly to uranium hexafluoride in a single step and therefore have great potential value for reprocessing irradiated fuels (to separate the uranium, plutonium, and fission products), as well as for initial production of uranium hexafluoride. In recent years, fused-salt processes that use either elemental fluorine or halogen fluorides to produce uranium hexafluoride and plutonium hexafluoride have been developed.[131,446]

Because uranium is radioactive and the hexafluoride is very volatile, strict control measures are applied in atomic energy installations and industrial plants in the United States that handle large amounts of the compound.[331] Emissions to the atmosphere from these sources are usually negligible. However, some accidental releases of uranium hexafluoride to the atmosphere have occurred.[79,298]

Test Firing of Rocket Engines with Fluorine and Fluorine Compounds

In recent years, test firings of rocket engines have been carried out with fluorine and fluorine compounds as oxidizers and with hydrogen, organic compounds, and nitrogen compounds as fuels. The combinations frequently react hypergolically–that is, ignite on contact–and have excellent propellant characteristics (high specific impulse and product of specific impulse and density). Although toxic gases, such as hydrogen fluoride, are formed in many of the reactions, the propellants are intended for use in second- or third-stage rockets, and the gases would therefore normally be released at high altitudes. Among the high-energy oxidizers that have been considered for use in rockets[29,215,217,309,310,628] and in some instances tested are liquid fluorine, mixtures of liquid fluorine and liquid oxygen, chlorine monofluoride (ClF), chlorine trifluoride (ClF_3), chlorine pentafluoride (ClF_5), bromine trifluoride (BrF_3), bromine pentafluoride (BrF_5), oxygen difluoride (OF_2), trioxygen difluoride in liquid oxygen ($O_3F_2-O_2$), nitrogen trifluoride (NF_3), and perchloryl fluoride (ClO_3F). Chlorine trifluoride has also been used as a hypergolic igniter for solid rocket propellants.[145]

In long-duration test firings of rocket engines with fluorine-containing oxidizers, the exhaust gases are passed through scrubbers to remove toxic products, as required by the National Aeronautics and Space Administration (NASA). However, some test firings lasting 2-4 sec are permitted without the use of scrubbers. The air-pollution-control measures used in test firings with fluorine-oxygen mixtures and fluorine compounds are described in McDonnell-Douglas Corporation and NASA publications.[58,572]

Other Industrial Uses of Fluorides

Solutions of hydrofluoric acid, ammonium bifluoride, and other fluoride salts are used as etching agents in the manufacture of quartz crystals for radiocommunication equipment. The etching process brings the crystals to the exact dimensions required for use as frequency-control devices in oscillator circuits. Gaseous hydrogen fluoride and silicon tetrafluoride are released during the process.

Mixtures of hydrofluoric acid and other acids, such as sulfuric and nitric acids, are frequently used in metal-pickling operations to remove rust and scale from metal surfaces. Iron, steel, nickel, aluminum, and many other metals are cleaned in rolling mills and factories by hot solutions of this type. Hydrogen fluoride is present in the air above the hydrofluoric acid–sulfuric acid vats; both hydrogen fluoride and nitrogen oxides are present in the air above the hydrofluoric acid–nitric acid vats.

In the electroplating industry, many types of fluorides are used in the metal-plating baths. Hydrofluoric acid, sodium fluoride, potassium fluoride, and ammonium bifluoride are used for electrodeposition of tin–nickel alloys. Fluorosilicic acid or fluoroboric acid (HBF_4) is often added to nickel- and lead-plating baths. Cyanide baths for plating copper, silver, or gold may contain potassium fluoride or ammonium bifluoride. And baths for the electrodeposition of cadmium–tin alloys may contain fluoroboric acid, ammonium fluoroborate, cadmium fluoroborate [$Cd(BF_4)_2$], and tin fluoroborate [$Sn(BF_4)_2$]. Fluorides are also used for electropolishing metals. Hydrofluoric acid is used in baths for electropolishing tantalum, and fluoroboric acid is used in baths for electropolishing stainless steel. Fumes from the solutions listed include hydrogen fluoride, silicon tetrafluoride, and boron trifluoride.

Hydrofluoric acid is also used in some types of dyeing, and sodium fluorosilicate is used by the leather-tanning industry, both as a brine solution for conditioning hides and as a powder for preserving dry hides.

Phosphors for cathode-ray tubes and fluorescent lamps are manufactured from many inorganic materials, including fluorides. One of the types now used in fluorescent lamps is made of synthetic fluorapatite [$Ca_{10}F_2(PO_4)_3$], activated with small amounts of manganese and antimony. The following fluorides have also been used for the preparation of manganese-activated phosphors: calcium fluoride, zinc fluoride (ZnF_2), magnesium fluoride (MgF_2), solid solutions of calcium fluoride and aluminum fluoride, and many complex fluorides ($NaZnF_3$, $KZnF_3$, K_2ZnF_4, Na_2MgF_4, $KMgF_3$, K_2MgF_4, $KCaF_3$, and $KCdF_3$).

PESTICIDES AS SOURCES OF FLUORIDE IN THE ENVIRONMENT

Sodium fluoride has been used as an insecticide for the control of cockroaches in the United States since about 1900. It has also been used to control poultry lice, cattle lice, grasshoppers, cutworms, earwigs, silverfish, and other chewing insects.[453] Pure sodium fluoride is white, but federal law requires blue or green coloring matter to be added to the compound in the United States so that it cannot be mistaken for flour, baking powder, or household items of similar appearance. The compound is highly toxic to humans when ingested in large amounts, and many persons have been poisoned by eating such foods as bread, pancakes, and puddings accidentally made with sodium fluoride instead of other ingredients.[267]

Several other inorganic fluorides have also been used as agricultural and household insecticides. Sodium fluorosilicate is used as a toxicant for grasshoppers, crickets, cutworms, and weevils in mixtures with bran and water and as a home mothproofing agent in dilute aqueous solutions. Barium fluorosilicate is used as a dust (25 wt %) and as a spray (3-8 lb/100 gal of water) to control chewing insects on plants. Cryolite is used as a dust (20-80%) and a spray (0.35-1.0%) to control the codling moth, Mexican bean beetle, flea beetle, and tomato worm. The barium compound has very low solubility in water (0.025 g/100 ml at 25 C) and is therefore less injurious to plants than other, more soluble fluorides.

The importance of inorganic fluorides as agricultural insecticides has greatly diminished in recent years, and the compounds now constitute a negligible fraction of all insecticides produced and used annually. The amount of sodium fluoride used as an insecticide by farmers in the United States in 1964, for example, was only 408,000 lb, in comparison with 7,156,000 lb of metal arsenates and 155,996,000 lb of total insecticides.[185] The amount of sodium fluoride used for this purpose in 1966 was only 6,000 lb, and the amount of sodium fluorosilicate was 3,000 lb. Barium fluorosilicate and cryolite were not used in significant amounts on farms in the United States in 1964 and 1966.[185,186]

Fluoride sprays used on fruit trees leave measurable amounts of fluoride on the fruits, even if several months elapse between spraying and harvesting. The residues can be removed from the surfaces of hard fruits, such as apples and pears, by washing the fruits with dilute hydrochloric acid and then water,[184] but they are more difficult to remove from soft fruits, such as apricots, plums, and peaches.

Many organic compounds containing fluorine have been found to be very effective insecticides.[130,453] These include fluoroacetic acid, which occurs naturally in some plants (see Chapter 7), fluoroacetamide, fluoroacetylphenyl urea, fluoroethyl acetate, 2-fluoroethoxyethyl-*N*,*N*-dimethyl carbamate, tetramethyl phosphorodiamidofluoridate, *n*-butyl-*n*-ethyl-α,α,α-trifluoro-2,6-dinitro-*p*-toluidine (Balan, Benefin), and 2,2-bis-(*p*-fluorophenyl)-1,1,1-trichloroethane. The last compound in this group is commonly called "DFDT," because it is the fluorine analog of DDT, 2,2-bis-(*p*-chlorophenyl)-1,1,1-trichloroethane. The compound is more expensive, and therefore used less frequently, than DDT, but it is superior to DDT in some respects. In comparison with chlorinated hydrocarbons and organic phosphorus compounds–now the most widely used types of insecticides–organic fluorine compounds have little importance as agricultural insecticides in the United States.[185,186]

Sulfuryl fluoride (SO_2F_2) has been used in a few instances as a home fumigant to destroy drywood termites.[453] Methyl sulfonyl fluoride (CH_3SO_2F) has also been used as a greenhouse fumigant to destroy ectoparasites.

Trifluoralin (α,α,α,-trifluoro-2,6-dinitro-*N*,*N*-dipropyl-*p*-toluidine) is widely used as an agricultural weed-killer (herbicide). In 1964, the amount of Trifluoralin used on farms in the United States was 735,000 lb;[185] this increased to 5,184,000 lb in 1966.[186] Fluometuron or Cotoran [1,1-dimethyl-3-(α,α,α-trifluoro-*m*-tolyl)urea] is also an effective fluorine-containing weed-killer, but it is not used to any appreciable extent in the United States.

Several inorganic and organic fluorine compounds have been used as rodenticides. These include sodium fluoride, sodium fluorosilicate, potassium fluorosilicate, ammonium fluorosilicate, potassium fluorophosphate (KPF_6), and sodium fluoroacetate (FCH_2COONa). The inorganic fluorides are not as widely used for this purpose now as in the past; they have been largely supplanted by organic phosphorus compounds and other types of rodenticides.

ADDITION OF FLUORIDE TO WATER, TOOTHPASTE, AND OTHER PRODUCTS

Fluoridation of water in the United States and Canada has been practiced since 1945, when the cities of Grand Rapids, Michigan; Newburgh, New York; Evanston, Illinois; and Brantford, Ontario, began adding fluoride chemicals to their water supplies to reduce the incidence of dental caries.[683] The combined population of the four cities was then less than 1 million, and at that time approximately 5 million persons in the United States also consumed water with appreciable natural fluoride content. Today, nearly 90 million persons in the United States use fluoridated water, and approximately 115 million use water low in fluoride.

The chemicals normally used for water fluoridation are sodium fluoride, sodium fluorosilicate, and fluorosilicic acid. In temperate climates, appropriate amounts of the chemicals are added to municipal water supplies to bring the total fluoride concentration to approximately 1 ppm by weight. This has been shown to be the optimal concentration for reducing dental caries without causing mottling of tooth enamel.[112,290] In tropical climates, the fluoride concentration is adjusted to less than 1 ppm by weight, because larger amounts of water are consumed.

Although water is the principal source of fluoride in an average human diet in the United States, fluoride can also be ingested from other sources. It is now frequently added to toothpastes, tooth powders, chewing gums, mouthwashes, vitamin supplements, and mineral supplements. It is also available in tablet form and is present in some drugs and medications. Much of the available information concerning the intake of fluoride from such sources has been reviewed by Stookey.[635] Fluoride tablets generally contain 0.25–1 mg of fluoride; fluoride-supplemented vitamin preparations usually contain 0.5 mg of fluoride. Sodium fluoride is the usual additive. Mouthwashes containing 0.02% or 0.1% of fluoride as sodium fluoride have been used. Commercially available dentifrices may contain sodium fluoride, sodium monofluorophosphate (Na_2PO_3F), or stannous fluoride (SnF_2), in amounts sufficient to supply 0.1% fluoride. Ericsson and Forsman[196] have reported that 21–26% of the fluoride in a mouthwash may be retained in the mouths of 3- to 6-year-old children after use of the solution containing 0.05% sodium fluoride. When 4- to 6-year-old children used a dentifrice containing 0.1% of fluoride (as sodium monofluorophosphate or sodium fluoride), 25–33% of the fluoride was retained in the month. The retention was 6–12% in adults who used dentifrices containing sodium fluoride, stannous fluoride, or sodium monofluorophosphate. Hargreaves *et al.*[259] found that 90% of 3- to 6-year-old children swallowed less than 0.5 g of toothpaste per day; only 7% of the children swallowed 1–2 g, and this did not occur regularly with any of the children.

Large doses of sodium fluoride (20–200 mg/day) have been given orally to patients with some bone diseases, such as osteoporosis and Paget's disease, for periods of up to a year.[605,635] Among the beneficial effects reported are decreased urinary losses of calcium, decreased pain and discomfort, increased rates of bone accretion, and increased bone mineralization.

A number of organic fluorine compounds have also been used as drugs for the treatment of other diseases and metabolic disorders.[136,605] These include fluorosteroids (fluorometholone, fluoxymesterone, fluocinoline acetonide, fluorandrenolone, betamethazone-21-valerate, and many others), as agents for clinical treatment of leukemia, breast cancer, psoriasis, eczema, rheumatoid arthritis, and asthma; Capacin (3-fluoro-4-hydroxyphenylacetic acid), for treatment of hyperthyroidism; fluorine-substituted benzothiadiazines (flumethiazide, hydroflumethiazide, bendroflumethiazide, polythiazide, and benzylhydroflumethiazide), as diuretics and agents for treatment of high blood pressure; and fluorine-substituted phenothiazines (trifluoropromazine, trifluoroperazine, and fluophenazine) and 4-fluorobutyrophenones (trifluperidol, haloperidol, fluanisone, methylperidol, benzoperidol, dehydrobenzoperidol, spiroperidol, and fluropipamide), as tranquilizers.

3

Environmental Fate and Transformation of Fluoride

TRANSFER OF FLUORIDE IN THE ENVIRONMENT

A simple summary of the transfer of fluoride in the environment is depicted in Figure 3-1, where the various routes by which fluoride passes to and from the atmosphere, hydrosphere, lithosphere, and biosphere are illustrated. Both the natural pathways and those due to man's activities are shown, but their relative magnitudes are not indicated.

Vulcanism and the entrainment of soil particles are the natural routes by which gaseous and particulate fluorides pass from the lithosphere to the atmosphere; vaporization and aerosol formation are possible routes from the hydrosphere. By deposition of particulate materials or through precipitation, atmospheric fluoride returns to the hydrosphere or lithosphere. The transfer of fluoride from the lithosphere to the hydrosphere can occur through the leaching of minerals or the addition of particulate matter to groundwater and streams. The return route would be by the deposition of sediments. The terrestrial portion of the biosphere is an intermediate stage in the passage of atmospheric fluoride to the lithosphere. Fluoride accumulated by vegetation can pass to the soil through

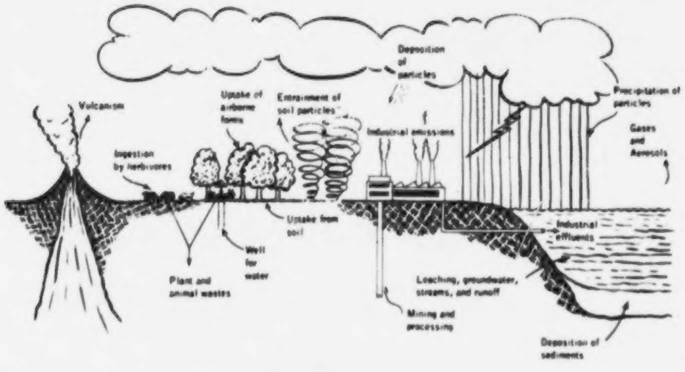

Deposition of particles
Volcanism
Uptake of airborne forms
Entrainment of soil particles
Industrial emissions
Precipitation of particles
Gases and Aerosols
Ingestion by herbivores
Uptake from soil
Well for water
Plant and animal wastes
Industrial effluents
Leaching, groundwater, streams, and runoff
Mining and processing
Deposition of sediments

FIGURE 3-1 Environmental transfer of fluoride.

plant wastes or enter the food chain through herbivores and ultimately through animal wastes. When fluoride is taken up by vegetation from the soil solution, its passage through the biosphere represents a recycling of lithospheric fluoride.

When industrial and other human activities are superimposed on the natural transfer mechanisms of fluoride, they result in new routes of transfer and an increased flow through pre-existing ones. For example, the emission of particulate and gaseous fluorides in the mining and industrial processing of fluoride-bearing minerals represents a path from the lithosphere to the atmosphere; this increased fluoride is removed from the atmosphere by natural means. The inhalation of fluoride by man and animals is another route into the biosphere, but is of main concern in industrial atmospheres. Similarly, the passage of lithospheric fluoride to the hydrosphere in industrial effluents and waste water will increase the fluoride burden in the aquatic environment and subsequent flow by natural routes to other phases of the environment.

There are no quantitative estimates of the amount of fluoride partitioned into the different phases of the environment, its residence times, or its rate of transfer. But a gross estimate of two pathways into the atmosphere can be obtained. It has been estimated that 30 million tons of soil are distributed into the atmosphere each year in the United States.[698] A mean fluoride concentration of 192 ppm implies about 6,000 tons of fluoride. Total emissions from industrial processes in the United States for 1968 were estimated at 119,000 tons.[686]

PHYSICAL AND CHEMICAL CHANGES OF AIRBORNE FLUORIDES

The range of dispersal of a fluoride in the atmosphere is determined not only by meteorologic conditions but also by the chemical stability and physical state of the fluoride (gas, aerosol, smoke, or dust). Many inorganic fluorine compounds are rapidly hydrolyzed by water vapor in the air and converted to less volatile compounds, which are removed from the atmosphere by condensation or nucleation processes. For example, silicon tetrafluoride, a major industrial pollutant, reacts with water vapor in air to form hydrated silica and fluorosilicic acid, as follows:[120,349]

$$3SiF_4 + 2H_2O \rightarrow SiO_2 + 2H_2SiF_6.$$

The same reaction occurs in aqueous scrubbers that are used to remove silicon tetrafluoride from factory off-gases.[620]

Fluorosilicic acid is highly soluble in water and is readily absorbed by plants.[106,281,282] In concentrated form (30% aqueous solution), it is moderately corrosive and attacks glass, ceramics, some metals, and metal oxides. The vapor phase above an aqueous solution of fluorosilicic acid contains both hydrogen fluoride and silicon tetrafluoride, the ratio of the first to the second decreasing with increasing concentration of fluorosilicic acid.[120,470] In the manufacture of wet-process phosphoric acid, the gypsum ponds in which waste liquids and solids are stored contain high concentrations of hydrofluoric acid and fluorosilicic acid. Liquid-vapor phase relations in the ponds, which are complex, are of some practical importance, in that the vapors of hydrogen fluoride and silicon tetrafluoride from the ponds constitute a major source of air pollution from this industry.[160,161]

Anhydrous hydrogen fluoride, which is emitted by many industries, combines readily with water vapor in the atmosphere to produce an aerosol or fog of aqueous hydrofluoric acid. Both the anhydrous and aqueous forms are easily absorbed by plant and animal tissues. The results of many plant fumigation experiments with hydrogen fluoride and other fluorides are described in Chapter 7.

The ability of hydrogen fluoride and aqueous hydrofluoric acid to etch glass is well known; frequently, the windows in glass factories (which use the acid), brickworks, and chemical factories bear witness to this. The reaction of hydrogen fluoride with glass yields silicon tetrafluoride, whereas the reaction of aqueous hydrofluoric acid yields fluorosilicic acid:

$$4HF + SiO_2 \rightarrow SiF_4 + 2H_2O;$$

$$nHF\ (aq.) + SiO_2 \rightarrow (n-6)HF\ (aq.) + H_2SiF_6\ (aq.) + 2H_2O.$$

Elemental fluorine (F_2) and strongly oxidizing compounds, such as some halogen fluorides (ClF, ClF_3, ClF_5, BrF_3, BrF_5, and IF_7), can produce injury in plant and animal tissues both by oxidation of the tissues and by deposition of fluoride ion after hydrolysis. Although fluorine and halogen fluorides are not common air pollutants, they are released in some rocket-engine test firings. The gases hydrolyze rapidly on contact with water vapor, forming hydrogen fluoride, oxygen, and other products. Fluorine hydrolyzes chiefly as follows:

$$F_2 + H_2O \rightarrow 2HF + \tfrac{1}{2}O_2.$$

A small amount of ozone is also formed. Hydrolysis reactions of the halogen fluorides are more complex and yield a variety of products, including HF, O_2, OF_2, Cl_2, HCl, $HOCl$, ClO_2F, ClO_2, ClO_3F Br_2, HBr, $HOBr$, BrO_2F, BrO_2, Br_2O, BrO_2F_3, I_2, HI, IOF_5, IO_2F, and IO_3F.[570]

Instrumental methods have recently been developed for measuring fluorine and chlorine trifluoride in the atmosphere at concentrations of 0-1,000 ppm and 0.01-1.0 ppm, respectively.[328,751] The methods can probably also be applied to the measurement of other very reactive fluorine-containing gases in the atmosphere.

Metal and nonmetal hexafluorides—such as hexafluorides of sulfur, selenium, tellurium, molybdenum, tungsten, platinum, and uranium—have various degrees of sensitivity to moisture. Sulfur hexafluoride (SF_6) is highly inert and is unaffected by water vapor at ordinary temperatures. Uranium hexafluoride, however, hydrolyzes rapidly to nonvolatile uranyl fluoride (UO_2F_2) and hydrogen fluoride, as follows:

$$UF_6 + 2H_2O \rightarrow UO_2F_2 + 4HF.$$

Although sulfur hexafluoride is nontoxic and is widely used as a dielectric medium in transformers, circuit breakers, and other electric devices,[112] several lower fluorides of sulfur are known (SF_2, S_2F_2, SF_4, and S_2F_{10}), of which at least two (SF_4 and S_2F_{10}) are extremely toxic.[184,235,333,614] Some of the lower fluorides are formed by the partial decomposition of sulfur hexafluoride in high-voltage electric discharges.[109] Sulfur tetrafluoride (SF_4) and disulfur difluoride (S_2F_2) hydrolyze readily, whereas disulfur decafluoride (S_2F_{10}) is unaffected by water. The hydrolysis reaction of the tetrafluoride forms thionyl fluoride (SOF_2) and hydrogen fluoride, as follows:

$$SF_4 + H_2O \rightarrow SOF_2 + 2HF.$$

Chlorofluorocarbon compounds of low toxicity—such as trichlorofluoromethane, dichlorodifluoromethane, and 1,2-dichlorotetrafluoroethane ($CClF_2CClF_2$)—are now widely used as aerosol propellants and refrigerants. Although the compounds are inert and not easily hydrolyzed, they decompose at high temperatures to toxic products.[182] If hydrogen and oxygen are present (from water vapor, air, and fuels), the toxic products frequently include hydrogen peroxide, hydrogen chloride (HCl), and phosgene ($COCl_2$). The decomposition can occur in furnaces and incinerators, on the burners of stoves, and on the hot filaments of electric appliances and can cause significant hazards to health if the toxic substances are not removed by adequate ventilation. Fluorocarbon plastics, such as polytetrafluoroethylene (Teflon) and polyhexafluoropropylene, also produce toxic substances when incinerated and therefore pose a similar health problem.

Most of the fluorides emitted by industries in the form of particulate matter are stable compounds that do not hydrolyze readily. Among the chief particulate emissions are cryolite, fluorapatite, calcium fluoride, aluminum fluoride, sodium fluoride, and sodium fluorosilicate. The solubility of each compound in water largely determines the availability of the fluoride in the compound to a plant or grazing animal. Calcium fluoride, for example, has very low solubility (0.0016 g/100 ml of water at 18 C) and is less readily absorbed by plants and animals than more soluble fluorides.[589] It has been shown that injuries to plants by soluble fluorides can be reduced by spraying lime solutions on the plants to precipitate calcium fluoride.[22] It has also been shown that injuries to cattle by fluorides can be reduced by adding aluminum sulfate to forage,[237] but no practical use appears to have been made of this method. Cryolite, aluminum fluoride, sodium fluorosilicate, and sodium fluoride have solubilities of approximately 0.039, 0.56, 0.65, and 4.22 g/100 ml of water, respectively, in the vicinity of room temperature. Fluorapatite is not very soluble in water, but it frequently causes fluorosis in sheep and cattle when it is present as dust on vegetation. The solubility in the gastrointestinal tract is probably enhanced by dilute hydrochloric acid. Climatic conditions are also important in determining the effects of particulate emissions on plants and animals.[504] Light rains may deposit fluorides from the atmosphere on vegetation, whereas heavy rains may wash off adherent fluoride dusts. In arid regions, the dusts may accumulate on vegetation over long periods and may be ingested in large amounts by grazing animals.

A recent study of effects of air pollutants on vegetation in the heavily industrialized Ruhr valley in Germany[326] produced evidence that fluorides and other air pollutants may act synergistically on plants. Plants in three areas were examined; in two of the areas, the plants were heavily damaged by fluoride emissions, but in the third area, the plants remained healthy, although exposed to similar concentrations of atmospheric fluorides. The phytotoxicity of the fluorides was reduced by emissions from a metal foundry that deposited with the fluorides on plants in the third area. The authors concluded that alkaline air pollutants, such as metal oxides, decrease the solubility—and therefore the phytotoxicity—of fluorides. They also indicated that some acidic air pollutants, such as sulfur dioxide, may have the opposite effect.

REGIONAL AND LOCAL VARIATIONS IN CONCENTRATIONS OF AIRBORNE FLUORIDE: EFFECTS ON THE ENVIRONMENT

In rural areas free of industrial or other types of contamination, the concentration of fluoride in air is extremely low. Usually it is measured in parts per billion or in micrograms per cubic meter of air (1 ppb by volume = approximately 0.8 $\mu g/m^3$). Very sensitive methods are required for analysis of the fluoride, and frequently no distinction is made between gaseous and particulate forms, which are collected and analyzed together. Of more than 7,700 air samples from many localities in the United States that were analyzed for total water-soluble fluoride by the National Air Pollution Control

Administration (NAPCA) in 1966 and 1967, 97% of those from nonurban areas had no detectable fluoride.[738] The highest concentration found in a nonurban location was 0.16 μg/m^3. In air samples from urban areas, the concentration of fluoride was less than 0.05 μg/m^3 (the lower limit of detection) in 87% of the samples and ranged from 0.05 to 1.89 μg/m^3 in the remainder of the samples. Higher levels of fluoride contamination than those reported here occur from time to time in localized areas close to some industries. However, the NAPCA measurements indicate that the normal concentrations of fluorides in air in both urban and rural locations are generally far below the concentrations known to be harmful to man.

The injurious effects on vegetation of fluoride emissions from brickworks, metal smelters, chemical manufacturing plants, fertilizer factories, and glassworks were noted in Europe during the last century, and reports of the injuries began appearing in the literature about 1870. Reports of injuries to animals from fluorides began to be published about 1930. Both natural and industrial sources of pollutants were shown to be capable of causing injury. In Iceland, illness of sheep after volcanic eruptions had been noted from very early times (about AD 1000), and Roholm[550] was able to diagnose the illness as fluorosis by analysis of bone specimens. A chronic illness of sheep and cattle in phosphate-rich regions of Algeria, Tunisia, and Morocco, known locally as "darmous," was shown by Velu[692,693] to be due principally to fluorapatite dust blown onto vegetation by prevailing winds. During the rainy season, the vegetation was washed free of the dust, and the condition of the animals improved markedly. Fluorosis of grazing animals from natural fluoride dusts and from industrial emissions has also been observed in the United States (see Chapter 8).

The effects of emissions from the phosphate mining and manufacturing complex near Bartow, Florida, have been studied in some detail. Here, huge draglines are used for open-pit mining of phosphate rock, producing fluorapatite dust, and gaseous and particulate fluorides are also liberated by numerous factories that produce phosphate fertilizers, elemental phosphorus, and phosphoric acid. Hendrickson[267] reported fluoride concentrations of 0–200 μg/m^3 in air samples of the region analyzed before 1959. Initially, a dynamic sampling method was used, in which fluoride was collected from the air at six sites on a variable schedule. A "limed-paper" sampling method was later adopted (Chapter 4), and the chemically treated papers were exposed for 1-month periods at approximately 40 sites in the area. From data obtained during March, June, September, and December 1959, Hendrickson prepared concentration isopleths showing the amounts of airborne fluoride collected by the papers at various distances from the industrial zones.[267] The highest concentrations of airborne fluoride were observed during the fall and winter. Huffstutler[299] recently reported fluoride concentrations of 0–42.1 ppb in air samples of the region analyzed during the years 1964–1969 (total water-soluble fluoride in 4-hr samples) and also reported fluoride concentrations in local pasture grasses and citrus plants. However, no correlation was noted between the concentrations in air and the concentrations in the grasses and plants.

Aluminum factories have been cited as sources of damaging fluoride emissions in Italy, France, Germany, the Soviet Union, Great Britain, Switzerland, the United States, and other industralized countries. One of the most thorough studies of the effects on man and domestic animals of emissions from aluminum factories was made by the Medical Research Council of Britain in the vicinity of Fort William, Scotland, during and after World War II.[17] Two aluminum-reduction plants were in the area, a large one at Fort William and a smaller one at Kinlochleven, about 10 miles away. During the war, aluminum was in great demand for aircraft production, and manufacture of the metal was rapidly expanded without great attention to safety aspects and working conditions. The findings of the Council therefore present a picture of an extreme case, in which emissions were not controlled and workers in the factories were exposed to concentrations of fluorides greatly in excess of those permitted by safety regulations today.

In the furnace rooms of the Fort William plant, very high concentrations of fluoride were found in air samples. A maximum of 3.6 mg/m^3 was found in one furnace room. Outside the factory, the concentration of fluoride in air dropped from approximately 0.22 mg/m^3 at a distance of 200 yards to 0.042 mg/m^3 at a distance of 1 mile (those were typical values during periods when the average furnace-room concentration was 1.42 mg/m^3). The fluoride emissions were deposited on soil and grass chiefly to the southwest and northeast of the factory, in the directions of the prevailing winds. Vegetation was destroyed for a distance of about a mile to the northeast, exposing a black surface layer of peat. Sheep and cattle in nearby pastures developed marked symptoms of fluorosis, such as "dagger teeth," which prevented normal mastication, and other deformities. However, persons living in the affected area (in Fort William, Glen Nevis, Inverlochy, Lochy Bridge, and Banavie) showed no signs of injury from the factory emissions other than a slight increase in the incidence of mottled teeth. Of 373 schoolchildren in the area, 21 (5.6%) had mottled teeth when examined dentally, but mottling was also observed in 3.1% of the schoolchildren of Moray and Nairn, communities east of Inverness-shire, in a later dental survey.

In the United States, the effects of fluorides on vegetation and cattle in the vicinity of aluminum plants along the Columbia River in Washington and Oregon—particularly on Sauvie Island, Oregon—have been studied by several groups of investigators.[135,636,684] Savara *et al.*[567] estimated that approximately 7,000 lb of fluoride per day was vented to the atmosphere by one aluminum plant near Vancouver, Washington, when it began operating in 1941 without any emission controls. Since that time, scrubber devices have been installed and have greatly reduced the emissions. Udall and Keller[684] examined the herds on Sauvie Island in 1950 and concluded that their poor condition was due to fluorosis. However, an investigating team from Washington and Oregon State Colleges analyzed the forage on the island in 1952 and 1953, studied the teeth, bones, and general condition of the cattle, analyzed urine samples, conducted postmortem examinations, and concluded that the cattle were not suffering from fluorosis.[636] From the conflicting reports, it seems probable that injury to cattle occurred to some extent during the early period when the aluminum plants were operated without adequate controls, but that very little occurred after about 1950. There were no instances of fluoride injury to persons living on the island. Savara *et al.*[567] conducted a dental and radiologic survey of 68 children living on the island and found no increase in the incidence of mottled teeth, no reduction in caries, and no changes in bone structure in x-ray pictures of the children's hands, wrists, elbows, and knees that would be indicative of the presence of excess fluoride.

A field study of the ponderosa pine blight area near Spokane, Washington, by a team of chemists, plant pathologists, entomologists, and foresters from the State College of Washington and the U.S. Department of Agriculture in 1949–1951[13,14] established that the tree injuries were caused by fluoride emissions from one or more industries in the area. At the time of the investigation, an aluminum-reduction plant, an oil refinery, and a smelter for magnesium and ferroalloys were operating in the district. Many trees had been killed in a 3-square-mile area around the industrial zone, and many more had turned brown in an outlying 50-square-mile area. Fluoride concentrations ranging from 0 to 351 ppb were found in air samples from 12 sites in the district in 1949 and 1950[13] (total water-soluble fluoride in air samples collected during 4-hr periods). The highest concentrations were found in sites where the greatest tree damage had occurred. In 1951, gladiolus, which is very sensitive to airborne fluoride, was planted at three sites in the area, and injury to the leaves was determined in July and August; concentrations of fluoride in air were also measured at the three sites. There was a close correlation between leaf injury and fluoride concentration at each site.[14] The experiment confirmed the earlier finding that fluoride was the chemical agent producing tree damage in the area.

On July 26, 1958, a new aluminum factory began operating west of the Columbia River near The Dalles, Oregon. This is a rich agricultural region with many orchards and farms producing cherries, peaches, apricots, alfalfa, and other crops, and the effects of fluoride emissions from the factory on the fruit trees and crops have been carefully observed. Reports issued periodically by the Agricultural Experiment Station of Oregon State University[150,152,154,155] present a chronologic record of the amounts of fluoride found in the air, foliage, and crops at various localities in The Dalles area. Environmental studies have also been conducted in the vicinity of Corvallis, Oregon, which has been considered a control area, relatively free of fluoride pollution. Before the start of operation of the aluminum factory in July 1958, the average fluoride content of samples of foliage from cherry and peach trees in The Dalles area did not exceed 13 ppm on a dry-weight basis (samples collected in 1953, 1957, and early 1958). No trend was noticed in the distribution of fluoride among orchards in different localities within the area, and the fluoride content of foliage was very similar to that in the Corvallis area. After the factory began operating, the average fluoride content of foliage of cherry and peach trees jumped to 65 and 76 ppm, respectively, in samples collected in October 1958. The highest average values (196 and 186 ppm, respectively) were recorded in September 1960, but the fluoride content in foliage has since dropped appreciably. Leaf scorch on apricot trees was observed in 1962 in many of the orchards and was most severe on the weaker trees.[154] Fluoride in alfalfa samples collected in July 1962 ranged from 6.5 to 83.6 ppm on a dry-weight basis (average, 31.2 ppm), and in samples collected in October 1962, from 53.0 to 166 ppm (average, 80.9 ppm). A seasonal trend was noted in the fluoride concentrations in all foliage and crops, with increases from summer to fall. Orchards and farms closest to the aluminum factory generally had the highest concentrations of fluoride in their foliage and crops. Ponderosa pine trees in The Dalles area were also affected. Pine needles collected within 1.8 miles of the factory contained 24–104 ppm of fluoride and showed various degrees of needle scorch.[150] Total fluoride concentrations in the air ranged from 0 to 12.9 $\mu g/m^3$ in samples analyzed in 1963 and 0 to 2.5 $\mu g/m^3$ in samples analyzed in 1964. In 1965, air samples were collected from five stations in the area for 12-hr periods, and in 1966 they were collected from 10 stations for 24-hr periods. They were again collected from 10 stations in 1967 for 6-, 12-, or 24-hr periods by automatic devices. In April of 1965, 1966, and 1967, the highest fluoride concentrations measured were 3.31, 4.77, and 4.65 $\mu g/m^3$, respectively. The concentrations dropped rapidly with distance from the factory within a radius of about 3 miles, but dropped very little thereafter. A good correlation was found between the concentration of fluoride in air and that in foliage in The Dalles area.[150] For the most part, high concentrations of fluoride in foliage were found only in samples collected within 3 miles of the aluminum factory, and amounts in samples collected beyond that distance were similar to those in the foliage before operation of the factory.

Schneider[574] recently published the fluoride concentrations found in air at a number of sites in the industrial city of Duisburg, Germany, and the surrounding area. The district contains steel mills, forges, metal smelters, and other types of heavy industry. Of a total of 1,339 air samples analyzed in 1965 and 1966, 90% had concentrations of fluoride between 0.5 and 3.8 $\mu g/m^3$, and the mean of all the measurements was 1.3 $\mu g/m^3$. Some very high concentrations (32 and 26 $\mu g/m^3$) measured in December 1963 and January 1966 were ascribed to emissions from a central heating plant. After the winter heating period of 1965–1966, most of the fluoride concentrations in the vicinity of the plant were in the range of 2–5 $\mu g/m^3$, although one high concentration, 16 $\mu g/m^3$, was again recorded.

In several of these studies, effects of meteorologic conditions on the distribution of airborne fluoride have been noted, but the observations have been too general to permit quantitative relationships to be established. Compton and co-workers[150,154,155] noted that the highest concentrations of fluoride in air and foliage occurred downwind from the aluminum factory producing fluoride emissions in The Dalles area. However, prevailing winds were judged by wind roses, and daily observations of wind velocity were not reported. In the study of emissions from the phosphate mining and manufacturing operations in central Florida, Hendrickson[267] found that maximal concentrations of fluoride in air occurred in winter, when the air was convectively stable and phosphate production was high, and that minimal concentrations occurred in summer, when the air was unstable and phosphate production was low. Adams *et al.*[14] reported wind conditions during periods of air sampling in the tree-damage area near Spokane, Washington; the highest concentrations of fluoride were found in air samples collected during periods of little or no wind. Thermal gradients and general observations of vertical air stability were not reported.

If the data permit any conclusions to be drawn on dispersion characteristics of fluorides, these are consistent with the general principle that fluoride—like other gases, liquids, and solids suspended in air—is dispersed downwind from the emitting sources and that it is dispersed vertically to a greater extent during periods of convective instability than during periods of inversion.

Several episodes of severe air pollution have been recorded in which many human fatalities occurred and in which fluorides from factories were present, as well as other types of pollutants. The lethality of fluorides in these episodes has never been established with any certainty. One episode of this type occurred in the Meuse River Valley in the region between Huy and Liege, Belgium, during the period December 1–5, 1930. A heavy fog laden with industrial wastes descended on the river valley and remained there for more than 4 days, reaching its greatest density on December 3 and 4. By the time the fog dispersed on December 5, several thousand persons had become ill and 60 had died. Industries in the area included several large steel mills and metallurgic plants, electric-power generating stations, glassworks, limekilns, zinc-manufacturing plants, a coking plant, a sulfuric acid factory, a fertilizer factory, and a gunpowder factory. In the official report of the tragedy,[205] sulfur dioxide and sulfuric acid aerosol were considered to be the most probable causes of illness and death. However, the report also listed many other chemicals as possible or probable emissions from the plants [H_2, SO_2, HCl, HF, NH_4F, ZnF_2, CO, CO_2, H_2S, NO, NO_2, HNO_2, HNO_3, $(NH_4)_2S$, ZnO, As_2O_3, AsH_3, $ZnCl_2$,

SiO_2, cement dust, lime dust, metal dusts, methane, ethane, unsaturated hydrocarbons, gasoline vapors, tar droplets, phenol, naphthalene, formaldehyde, methyl alcohol, and ethyl alcohol]. Although Roholm[552,553] and others[234,690] have suggested that fluorides contributed significantly to the disaster, most of the evidence indicates that other toxic agents, or combinations of agents, were responsible. Two members of the official investigating committee have elaborated their reasons for not considering hydrofluoric acid to be a primary cause of the deaths and injuries.[405,406]

BIOACCUMULATION AND ORGANIC TRANSFORMATION OF FLUORIDE

Atmospheric fluorides are absorbed and accumulated by leaves (see Chapter 7). The ingestion of fluoride-containing material by animals also results in a steady accumulation of fluoride within the animal body. This fluoride accumulates almost exclusively in skeletal tissues; the nature of this accumulation will be discussed in detail in Chapters 8 and 9.

Several species of plants have the capacity to convert fluoride obtained from the soil or soil water into carbon–fluorine compounds that have a much greater mammalian toxicity than inorganic fluoride salts. In 1944, Marais[408] reported the presence of monofluoroacetic acid in the South African plant, *Dichapetalum cymosum*, or gifblaar. The related species *D. toxicarium* was found to synthesize ω-fluoro-oleic, ω-fluoropalmitic, and ω-fluoromyristic acids.[702] Subsequent studies have shown the presence of monofluoroacetic acid in *Palicourea marcgravii* in Brazil,[174] *Acacia georginae* in Australia,[491] and a number of species of *Gastrolobium* and *Oxylobium* in Australia.[34–39,435]

Recently, a group from Utah State University[138,378,736] has reported the presence of both monofluoroacetic and monofluorocitric acids in mixed pasture forage, crested wheat grass, and soybeans exposed to atmospheric fluoride. These findings have not yet been verified by other groups.

BIOLOGIC CHAINS

The biologic chains important in the consideration of fluoride as an atmospheric pollutant are from the atmosphere to humans or animals, from the atmosphere to plants, from the atmosphere to plants to humans or grazing animals, and from the atmosphere to plants to grazing animals to humans.

The quantitative relation between the concentration of fluoride in the ambient air and that absorbed by man and domestic animals will be considered in detail in the sections on the effects of fluorides on animal and human health (Chapters 8 and 9). It is clear, however, that this is a minor route of exposure for grazing animals, and that ingestion is their chief route of fluoride exposure. The edible portions of most crops grown for human consumption do not have high enough fluoride content from exposures to atmospheric fluoride to increase fluoride intake by humans significantly, and inhalation may be their major route of exposure.

Fluorides do accumulate in the skeletons of animals that consume high concentrations of fluoride. However, consumption of the edible portion of these animals by humans will add an insignificant amount of fluoride to that already present in the diet. There is adequate information to indicate that milk from animals fed high concentrations of fluoride is not sufficiently increased in fluoride to be a serious problem.

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Environmental Analysis of Fluoride

Fluoride injury to plants and animals presents a number of diagnostic problems, especially in the case of plants. For example, the various types of lesions produced on plants can be confused with those caused by diseases, insects, and physiologic disorders. Thus, a definite diagnosis of fluoride injury usually requires analyses of the fluoride content of the receptor. The ability to predict an effect on plants may require a knowledge of both short- and long-term atmospheric fluoride concentrations. Because both gaseous and particulate fluorine compounds are emitted by industry and the gaseous form is considered to be the more toxic to plant life, methods capable of distinguishing between these two physical forms of fluoride are required.

It is obvious, therefore, that establishment and implementation of air-quality standards create a demand for methods of sampling and analysis that are sensitive, accurate, and precise. A standard for atmospheric fluoride cannot be enforced if methods are not available to separate and collect gaseous and particulate fluorides or if the standard is below the minimal level of accurate detection. The same may be said for a forage standard.

It is important to emphasize and distinguish between these two interrelated criteria of analytic methods. Noncompliance is often suspected if a single determination exceeds a standard. Therefore, methods possessing both high reproducibility and accuracy are required in evaluating an air-pollution problem.

A recent collaborative study[315] of fluoride in vegetation indicated a large relative standard deviation among the 31 participants, ranging from 12.7% to 25.4%. That emphasizes the need for care in the various steps of fluoride analysis and for standardization of methods.[305,306]

SAMPLING AND SAMPLE PREPARATION

One is seldom interested in the fluoride content of a segment of the universe so small that the entire segment may be analyzed, but rather in the fluoride content of a pasture, of a municipal water supply, or of the blood of an exposed group of workers. The entire pasture, the entire water supply, or all the blood from the workers cannot be analyzed; one must be content with analyzing samples.

There are no hard and fast rules applicable to all sampling problems. Common sense and the recognition that a degree of variability among individuals is a characteristic of every population are the only guides. Each new study may require the development of a specific sampling scheme. Each sampling scheme should be designed to yield a valid estimate of the variability of the fluoride content among individuals in the population, as well as a valid estimate of the average fluoride content of the entire population. Benedict[64] has proposed a sampling scheme for forage samples.

After sampling, various preparative steps are required before the fluorides can be separated—sorting, screening, grinding, drying, ashing, alkaline fusion, extraction, evaporation, and so on.

Relatively simple, primarily inorganic systems (such as water) may not require sample preparation. Urine, blood, and other body fluids may also often be handled with a minimum of preparation. More complex organic systems generally require ashing or digestion either to free the fluoride from the matrix or simply to reduce the bulk. Some organic systems and many inorganic systems require fusion with alkali to free the fluoride from refractory complexes.

In all the preparative steps, contamination is an ever-present hazard, and the chance of contamination increases with the number of steps and the number and amount of reagents required.

Losses of volatile fluoride may occur during evaporation or ashing steps required to reduce sample bulk or during sample storage. Whenever digestion, ashing, or fusion is required to break down refractory compounds, losses due to volatilization or incomplete release must be considered.

Plant Tissue

For the analysis of total fluoride in plant tissue, samples must usually be dried, ashed, and fused with alkali before a separation step may be applied. These steps are now reasonably well standardized.[23,305,306,316,346,482,493,710,711]

The first step, however, is a mental one: the question of what is meant by "total fluoride." Are we interested only in the fluoride *in* the tissue or in the fluoride both *in* and *on* the tissue? Surface contamination from soil and dust, not only on root tissue but also on aboveground portions of the plant, is an old problem in plant analysis. It is especially troublesome in trace-element analysis, and no completely satisfactory procedure has appeared. The decision as to whether tissues should be washed will depend on the intended use of the analytic data. Thus, if one wishes to estimate the dietary fluoride intake of a cow, data representing the fluoride available for ingestion are important. If interest is directed to the response of the plant itself, data representing the fluoride in and not on the tissue are important.

Particulate fluorides from industrial dusts, soil, or other sources on the surface of a forage are consumed by animals. But the availability of fluorides to an animal system varies, and the amount that is available has not been clarified to the satisfaction of all. The analytic tools necessary to provide definitive estimates of availability of fluoride to animals are lacking.

WASHING

An estimate of the fluoride incorporated into plant tissue may require washing, but it is necessary to recognize that washing may not remove all surface fluoride and may extract fluoride from within the tissue. A great number of washing methods have been used. Most consist of swirling or rubbing the foliar tissues in an aqueous solution of soap or detergent for a defined or undefined period. It is obvious that consistent results among laboratories will require acceptance of a uniform washing procedure. The following method has been tentatively accepted by the Intersociety Committee on Methods for Ambient Air Sampling and Analysis:[306]

> Freshly harvested tissues are washed in a solution of 0.05 per cent Alconox* and 0.05 per cent Na_4 EDTA† in a polyethylene container for 30 sec with gentle agitation. The tissue is removed and allowed to drain for a few sec and is then rinsed for 10 sec in each of three beakers of deionized water.

STORAGE AND DRYING

Improper handling of samples can result in loss of fluoride during transport, storage, or drying. Unwashed or washed samples are commonly stored in the frozen state with calcium oxide added[418,482] to prevent loss of fluoride. Sometimes the plant tissue is oven-dried at 70–80 C for 24–48 hr without addition of a lime fixative.[242,306,416] Gwirtsman *et al.*[242] and Mavrodineanu *et al.*[416] present results of fluoride determinations of the same tissues analyzed fresh and after drying.

> Data from 95 samples representing seven different crops . . . show that the results obtained by the two procedures with comparable samples showed a correlation coefficient of 0.989. A routine procedure employing dried plant material for analysis has been preferred because of its convenience. However, the data . . . show that analyses of alfalfa (*Medicago sativa* L.) fixed when fresh resulted in significantly higher fluoride values than did analyses of comparable dried samples when control (nonfumigated) [plants] or plants fumigated with 8 p.p.b. of HF were examined. Fresh and dried materials yielded equivalent results when the alfalfa plants were fumigated with 17 p.p.b. of HF. The possible effects of dew, rain, or dust were not considered in these experiments.[416]

It appears that oven drying of samples at 70–80 C is satisfactory, although fixation with lime before drying or for cold storage may be preferred under some circumstances.

ASHING AND ALKALINE FUSION

The ashing step is usually carried out in nickel, Inconel, or platinum crucibles. Temperatures of 400–600 C for about a 2-hr ashing are commonly used in the United States;[242,306,416,482,711] some European workers ash to temperatures of 450 C and above 600 C.[23] Thomas and Amtower[672] recommended ashing by oxygen-flask combustion.

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*Alconox Inc., New York, N.Y.

†Tetrasodium salt of ethylenediaminetetraacetic acid.

Remmert *et al.*[535] and Rowley *et al.*[560] were among the first to recognize that rather refractory fluoride compounds were formed during ashing of many plant samples–particularly of high-silicate species, such as the grasses. Fusion of the ash with alkali (usually sodium hydroxide pellets[306] or an ethanolic solution of sodium hydroxide[305]) has therefore become a standard practice.[23,242,286,288,346,416,482,560,711] Both the lime used to fix volatile fluoride compounds during drying, storage, and ashing and the alkali used in fusion are potential sources of contamination. Thus, these reagents must be selected with care and should be included in the blank determinations.[286,288]

After fusion, the cooled melt is solubilized, and separation and estimation of fluoride are carried out by suitable procedures.

Animal Tissue

STORAGE, DRYING, AND ASHING

Most animal tissues may be stored in the frozen or oven-dried state. The ashing procedure analogous to the method for plant tissue is probably the most accurate. Although there are no data to support the need for lime or alkali during drying and ashing, they are commonly used to reduce possible loss of fluoride by volatilization. Pool *et al.*,[524] in their analysis of chicken meat, added calcium hydroxide before ashing, but produced no data or references demonstrating the need to do so. Because most soft tissue contains little silica or aluminum, alkali fusion of the ash is probably not necessary.

SOFT TISSUE

Generally, soft animal tissues are dried and ashed, and the fluoride is separated by distillation.[140,354,440,652] Some investigators have applied a diffusion-separation scheme directly to macerated soft tissue.[56,250,610] That scheme may not have been sufficiently investigated to be completely effective for all forms of fluoride in the tissue.[662] More recently, analyses of tissue fluoride content have been carried out by extracting the tissue with perchloric acid and determining the amount of inorganic fluoride by use of the specific ion electrode.[582]

SKELETAL TISSUE

Most of the fluoride in animal tissues is in the skeleton, analysis of which poses no problem. Bones can simply be cleaned to free them from flesh, dried, and then ashed at 500–600 C. The ashed sample can be added directly to a distillation flask in the manual steam-distillation method[283,578,642,647] or to a diffusion dish.[250,599] The ash can also be dissolved in a small amount of acid for determination by a semiautomated method or for determination by use of the specific ion electrode.[597] Ion-exchange procedures have also been used[486] for skeletal tissue, and similar procedures can be used to determine the amount of fluoride in tooth samples. Some investigators have also used a diffusion–separation scheme for ground bone and tooth samples.[56,250,599]

The fluoride content of different bones of the skeleton varies appreciably[595] and there are significant changes in the fluoride content of different parts of the same bone.[223,647] It is therefore important for reports of the skeletal fluoride content of large animals to indicate which particular bones were analyzed and in what ways particular bones were sampled. Without such information, results can be extremely misleading.

BODY FLUIDS

Blood, serum, and urine are often evaporated, ashed, and then treated like any other dry samples. A number of direct methods of analysis, requiring little or no pretreatment, are coming into wide use.

Even when excessive amounts of fluorides are ingested, the fluoride contents of the plasma and soft tissues are extremely low, and their determination is difficult. Both microdistillation methods[596] and diffusion methods[156,599] have been published for the determination of plasma fluoride. Although significant increases in fluoride concentration have been seen in experiments in which large animals[124,594] or rats[583,593] were given relatively high doses of fluoride, it has been difficult to detect any increase in plasma fluoride concentrations in human populations where the water fluoride content was a variable.[600] There now appears to be agreement[601,662] that much of the fluoride present in human plasma is in a nonexchangeable form, and Taves[660,665] has published a method for the determination of the ionic, exchangeable form relative to the total amount of fluoride present in plasma or serum. Using this method, it is possible to determine that there are significant increases in plasma fluoride content in the human population where fluoride intake from the water supply is a variable or where patients have been given different amounts of sodium fluoride as a treatment for bone disease.[663] A method for the determination of plasma fluoride concentrations by use of the specific ion electrode has also been published,[53,212] and has now shown that fluoride concentrations determined by use of the electrode represent the exchangeable portions present in plasma. The nature of the so-called nonexchangeable fluoride has not been determined, but there are indications[661] that it is tightly bound to plasma proteins.

A number of procedures have been used for the determination of fluoride in urine. Fluoride in urine exists in the presence of relatively large amounts of substances that may cause serious interference in the common methods of measurement, and several techniques have been developed to separate the fluoride from such substances. These include distillation, ion exchange, and diffusion, after which the fluoride may be measured by visual titration or, more commonly, spectrophotometry. Urine samples can also be diluted, treated with silver perchlorate ($AgClO_4$), and subjected to direct analysis by a semiautomated method without prior ashing.[176]

Distillation procedures require the prior fixing of fluoride by the addition of a fixative, such as calcium oxide or magnesium salts, followed by ashing to destroy organic matter; the fluoride is then separated by steam distillation from a sulfuric or perchloric acid solution of the residue. Factors important to the fixing, ashing, and distillation have been reviewed by Cholak.[140] Ion-exchange procedures for urinalysis were introduced by Talvitie and Brewer[657] and Dubois *et al.*[181] who used a photometric procedure to measure the separated fluoride. Several investigators have described diffusion methods for the separation of fluoride from urine. Rowley and Farrah[201,559] discuss critical factors, and improvements in the technique are presented by Cumpston and Dinman.[162]

Methods have been developed for the visual titration of the separated fluoride, but they have been largely supplanted in routine analysis by spectrophotometry.

The advent of the fluoride-specific ion electrode has now made the measurement of urinary fluoride concentrations easy, rapid, and routine.[53,472,637,682] The device uses a crystal that responds only to the fluoride ion (F^-) and a calomel reference electrode. The potential due to F^- in a buffered urine sample is measured on an expanded-scale pH meter. The potential varies linearly with the negative logarithm of the fluoride concentration, and the millivolt readings obtained can be converted directly to micrograms of fluoride. Prior separation is not required, and the analysis can be performed in a few minutes. EDTA may be added to the urine after collection to prevent precipitation of insoluble compounds with phosphate and fluoride. The presence of citrate in the buffered sample while the fluoride potential is being measured is believed to prevent the formation of complexes of iron or aluminum that might otherwise remove F^- from the system.

Of the various procedures mentioned, separation of fluoride by fixing, ashing, and distilling followed by visual titration is the oldest. Although it is reliable in careful hands, its sensitivity depends to some extent on the ability of the analyst to detect subtle color changes. This can be obviated by spectrophotometry. Much of the delay in analyses arises during ashing of the sample, and the ion-exchange and diffusion procedures also specify an overnight interval for separation of the fluoride. In the spectrophotometric procedures, it must be remembered that the bleaching of the color developed, which is the basis for the quantitation of the fluoride, is not specific for fluoride. Interference by other agents, which may still be present, is eliminated, minimized, or controlled by adhering to rather empirically established conditions.

Use of the fluoride-specific ion electrode offers the most rapid, simple, and specific method for measuring fluoride, although even here there are limitations. At a pH below about 4.5, undissociated hydrogen fluoride forms and will not be measured, whereas at a pH above 8, the electrode also responds to hydroxyl ion. The high initial cost of the equipment may also be a difficulty, but this may be compensated for by the smaller demand on the analyst's time.

Water Samples

Water analysis has been fairly well standardized.[63,111,287,332,447,449] The direct use of the specific ion electrodes is gaining favor for monitoring of public water supplies.[50] Industrial waste waters present specialized problems, depending on the other materials present. Waters containing high concentrations of phosphate, silicates, and other fluoride-complexing materials, such as aluminum, require special precautions and procedures. The reference method for comparison of other procedures should involve evaporation, ashing, and fusion to ensure complete recovery of fluoride from possible refractory complexes. Some liquid samples may be steam-distilled directly.[176,454]

Soils, Minerals, and Miscellaneous Compounds

The preparation of soils and minerals often requires grinding in ball or hammer mills. Contamination is a hazard. The fluoride content in samples of interest is usually high (up to about 5%), so that trace contamination is usually less important than in biologic materials.

Samples (such as of soils) that may contain appreciable quantities of organic matter must be ashed. Most of the samples in this group must be fused with alkali.

The discussions on distillation by Fox and Hill[207] and Fox and Jackson[208] are generally applicable to all mineral samples. The general method of MacIntire[387] for soils is still adequate and applicable to a variety of minerals. Abernethy and Gibson[1] discussed methods for coal. Brewer[99] reviewed the problem and presented a scheme for all soils and minerals.

Air

Sampling the air for fluoride is subject to the same problems inherent in all the air-sampling techniques discussed by Hendrickson.[266] Atmospheric sampling for fluoride, however, is further complicated by the extremely low concentrations normally found in air and by the presence of both gaseous and particulate forms. Furthermore, because gaseous fluoride compounds are more toxic to vegetation than the particulate forms,[429,507] it is important that the sampling method used be capable of separating the two forms. Pack *et al.*[507] reviewed and assessed the sampling techniques available in 1960, and the later review by Farrah[202] added new information. The discussion on calibration techniques by Jacobson[313] is applicable to all methods.

MANUAL DYNAMIC METHODS

Total fluorides in both the gaseous and particulate forms have been collected with good efficiency in standard bubblers and impingers,[202,507] special bubblers,[242,414] silica-gel columns,[654] alkali-treated filter papers,[302] and glass-fiber filters.[506] Each of these systems is highly efficient for collection of gaseous fluorine compounds, but they vary somewhat in their efficiency for particulate fluorides, especially at particle sizes of less than 0.3 μm.[202,505–507] For both impingers and bubblers, no differences in efficiency have been found between water and dilute alkali as collection liquids.[202,507]

The glass-fiber filter suggested by Pack *et al.*[506] is a simple, rugged system with good efficiency for both gaseous and particulate fluorine compounds, but the method suffers from a fairly high blank value. Under some conditions, the filter may become saturated, with an accompanying loss in efficiency. However, for most purposes, this saturation limit (75 μg/in.^{2} for 5% loss)[505] is satisfactory.

The various bubblers and impingers are somewhat variable in efficiency for total fluoride,[507] but adequate for many purposes. The main deficiency in these systems is the maintenance of liquid level during long sampling times,[266] although suggestions for the solution of this problem have been published.[327]

For most samples, the liquid from the sampler and the filter paper or glass-fiber filter[302,506] can be introduced directly into a steam-distillation system.[202,305] When highly refractory dusts or soil particles are present, fusion with alkali is necessary.[202,305]

There are several methods for separating and collecting gaseous and particulate fluorine compounds. Some rely on the high reactivity of the gaseous forms (principally hydrogen fluoride and silicon tetrafluoride) with aluminum, alkali, and other materials. Pack *et al.*[507] have described the use of concentric aluminum tubes, Buck and Stratmann[114] have used tubes filled with silver or quartz beads coated with sodium carbonate, and Pack *et al.*[507] and Mandl *et al.*[407] have used bicarbonate-coated glass tubes for selective absorption of gaseous fluorine compounds in the presence of particulate matter. The latter may then be collected in an impinger or on an appropriate filter. Pack *et al.*[507] also reported that membrane filters are efficient collectors of particles and have a low reactivity with gaseous forms, allowing them to pass through the filter to be collected by other means. Habel[244] refined that method by using a membrane filter in a heated quartz filter holder in series with a collection device. Mandl *et al.*[407] have recently described a paper-tape sampler that accommodates two treated-paper tapes, one to remove particulate and the other to remove gaseous fluorine compounds.

In the case of concentric aluminum tubes,[507] the gaseous fluorine compounds are removed by reaction with aluminum, and then the particles are collected. Because the reacted fluoride cannot be easily removed, the concentration of gaseous fluoride is estimated on the basis of the difference between the particles collected after removal of the gaseous component and a parallel sampler collecting total fluoride. Pack and Hill[505] reported an efficiency of 96% or better for collection of hydrogen fluoride on aluminum.

Bicarbonate-coated glass tubes have the advantages of simplicity, ease of removal of the coating for analysis, and a reasonably high degree of separation of gaseous from particulate fluorine compounds. Thus, Pack *et al.*[507] have reported a collection efficiency for hydrogen fluoride of up to 90% using glass tubes 10 mm in inside diameter and 3 or 4 ft long and retention on the tube of less than 8% of a "large" aerosol (80% larger than 2.0 μm) and about 4% of a "small" aerosol (70% smaller than 2.0 μm). Mandl *et al.*[407] have shown that the air-sampling rate and tube diameter are important dimensions for efficient collection of hydrogen fluoride. They found that coated glass tubes 10 mm in inside diameter and 4 ft long were between 66% and 89% efficient for hydrogen fluoride, with efficiency being inversely related to air flow rates. Apparent uptake of micronized cryolite (mean diameter, 1.5 μm) on the coating was 6% or less. Higher collection efficiencies for hydrogen fluoride were obtained with tubes 5 and 7 mm in inside diameter and at correspondingly lower air flow rates. Mandl *et al.*[407] recommended the following: a bicarbonate-coated tube, 7 mm in inside diameter, 4 ft long, with an air-sampling rate of 0.5 cfm or less, with particles collected on a membrane filter mounted at the outlet end of the tube.

Membrane filters of appropriate pore size are highly efficient collectors of particles, and some have a relatively low reactivity with gaseous fluoride. Pack *et al.*[507] reported a collection efficiency of better than 98% for both "small" and "large" aerosols (70% smaller than 2.0 μm and 80% larger than 2.0 μm, respectively) and retention of only about 7% of hydrogen fluoride on a membrane filter. However, the type of membrane filter used is very important, in that some will collect significant amounts of hydrogen fluoride. Mandl *et al.*[407] have reported that treatment of the filter with a solution of citric acid significantly reduced the retention of hydrogen fluoride. Farrah[202] emphasized that special care and treatment of the filter holder and support for the membrane filter are necessary to avoid adsorption of gaseous fluorine compounds. After enclosing the membrane filter in a heated quartz filter holder, Habel[244] reported excellent retention of particles and quantitative passage of gaseous fluorine compounds.

The principle of using a prefilter that removes particles and allows passage of gaseous fluoride was incorporated in a double paper–tape sampler by Mandl *et al.*[407] The air sample passes in sequence through a Whatman No. 52 paper tape treated with citric acid and a Whatman No. 4 paper tape impregnated with sodium hydroxide. Particles are retained on the citric-acid-treated tape, and gaseous fluoride reacts with the alkali-treated tape. Retention of hydrogen fluoride on the prefilter ranged from 0 to 5.8%, averaging about 1% for all tests.

AUTOMATIC DYNAMIC METHODS

The low concentration of fluorine in the ambient air has severely limited the development of instruments capable of providing a continuous record of atmospheric fluoride concentration over relatively short periods.

Adams and Koppe[10] described a dose-type automatic instrument, which is commercially available as the Mini-Adak. In one form, the instrument used the zirconium–Eriochrome Cyanine R reagent, although the SPADNS* reagent is now in general use. In a modification for recording high levels in stack gases, the iron–ferron complex reagent was recommended.[3] The reported minimal detection limit with zirconium–Eriochrome Cyanine R is about 0.25 μg of fluoride.[10] The sampling rate has been designed for 1 cfm and a cycle of up to 5 hr, although the commercial model uses a considerably lower sampling rate. Thus, for a 3-hr sampling cycle, the detection limit would average approximately 0.05 $\mu g/m^3$ for 3 hr. In their field test of the instrument,[9] the authors suggested a practical detection limit of 0.4 $\mu g/m^3$ if present for 20–30 min. For many monitoring applications, this sensitivity may be adequate. The instrument, however, has one serious limitation: It measures the gaseous fluorine component plus some portion of the particulate component, depending on its composition. Tests with cryolite aerosol[12] indicated an efficiency of about 18% for this material.

Ivie *et al.*[308] and Thompson *et al.*[676] have reported on prototypes of a sensitive automatic instrument for hydrogen fluoride. These are based on a patent[132] and an earlier prototype described by Thomas *et al.*[674] The detection system is based on the quenching by hydrogen fluoride of the fluorescence of the magnesium salt of 8-hydroxyquino-

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*Sodium 2-(*p*-sulfophenylazo)-1,8-dihydroxynaphthalene.

line with which a paper tape is impregnated. Two air streams are drawn through two glass tubes, one of which is coated with sodium bicarbonate to remove gaseous hydrogen fluoride. The air streams are passed through adjacent spots of the chemically treated paper tape. The presence of hydrogen fluoride quenches the normal fluorescence of the treated paper under ultraviolet light. The difference in emitted light between the two areas of the tape is monitored by reflecting the two beams of light onto photomultiplier tubes, with the differential output being transmitted to a recorder.

Data presented in the description of the most recent version of this instrument[676] indicate a sensitivity to less than 0.1 $\mu g/m^3$ with a 30-min cycle.

The fluorescence-quenching method has several desirable features: good sensitivity, specificity for hydrogen fluoride and not for particulate fluorine compounds, and dry collection and detection. Unfortunately, however, final development has been beset with electronic problems and sensitivity to atmospheric moisture, and no model is commercially available.

Taylor and Sefton[579,667] have described an automatic carbonate-coated tube analyzer, which operates in the following sequence: A solution of sodium carbonate and glycerol is pumped through the sampling tube, and ambient air is drawn through the tube for a preset period; at the end of the sampling period, the tube is flushed with zirconium–Eriochrome Cyanine R reagent, and the absorbance of the sample is determined and recorded; the cycle is then repeated. Air flow can be adjusted to provide sampling periods ranging from 15 to 150 min. The dead time between cycles is between 2 and 3 min. The authors reported a minimal detection of 0.1 $\mu g/m^3$ with the maximal flow rate (30 liters/min) and the longest sampling time (150 min). The upper limit for these conditions is about 8 $\mu g/m^3$. The commercial models, however, provide a sampling rate of about 20 liters/min.

For the high levels of hydrogen fluoride found within industrial plants and in stack gases, various other instruments have been suggested. The modification[3] of the instrument developed by Adams and co-workers has been noted. Howard and Weber[297] have suggested an instrument based on an aluminum–platinum electrode.

STATIC METHODS

The establishment of a comprehensive atmospheric sampling network may prove to be impractical from an economic standpoint. Automatic monitors, of course, represent a greater initial investment than manual samplers. However, because the latter devices collect but do not analyze fluorides, they represent an increased analytic load. Therefore, static monitoring methods may play an important role in area surveillance, where one is interested in the relative atmospheric contamination at a number of different sites.

Vegetation (particularly grasses and other forage plants) has often been used as a static monitor for atmospheric fluorides.[13,80,144,151,153,175,269,293,318,319,581,621,658,718] The accumulations of fluorides in vegetation at different times may be used as relative estimates of the degree of fluoride pollution. The use of plants as monitors, however, presents several problems: errors in collecting representative samples of vegetation for analysis, distribution of a particular plant species over a wide area, and seasonal differences in growth. Weinstein and McCune[714] have reviewed the procedures and problems of vegetation sampling and discussed other uses of plants as biologic monitors.

Several static methods have been suggested as substitutes for estimating the uptake of fluorides by vegetation. Ost[499] hung lime-impregnated cloth in trees and attempted to relate the amount of fluoride found on the cloth to the amount found in nearby vegetation. MacIntire *et al.*[389] suggested using Spanish moss as a static monitor. Miller *et al.*[458] proposed using filter paper impregnated with a calcium salt. The "limed" papers were exposed in a shelter to protect them from rain for several weeks (usually 4), and then collected and analyzed for fluoride. The authors reported reasonably good correlations between the amount of fluoride taken up by the filter papers and the amount of fluoride in gladiolus, the amount of tip necrosis on gladiolus leaves, and the amount of fluoride in forage crops. In an area of low atmospheric fluoride, the correlation between amounts of fluoride in limed filter papers and in forage was not as good as in areas of greater pollution. The technique has had considerable field use,[4,5,458,547,726] and Adams[5] has provided information relating uptake of fluoride on limed papers to the fluoride content of air in controlled fumigation chambers.

The limed-filter-paper method is relatively simple, inexpensive, and useful in defining potential areas of hazard to vegetation when used in conjunction with other estimates.

SEPARATION OF FLUORIDES

Most of the methods for estimating fluoride concentrations are sensitive to interfering substances. For most analytic applications, therefore, a separation step is required to isolate fluoride from those substances.

Precipitation

Precipitation methods are generally not in use as a means of separating fluorine from interfering substances, although they have been used in the gravimetric method of determination (mentioned below).

Ion Exchange

J.P. Nielsen and Dangerfield[487] suggested the use of anion-exchange resin in the hydroxyl form as a means of concentrating the fluoride collected in bubblers or impingers. H.M. Nielsen[485,486] suggested a more general procedure using the acetate form of an anion-exchange resin and applied it to some simple ashed biologic materials as a concentrating step after standard distillation. Some modifications in this procedure were made by Newman.[478].

Talvitie and Brewer[657] used a cation-exchange resin in the hydrogen form to preserve urine. The diluted urine was passed through an anion-exchange resin in the hydroxyl form and eluted with beryllium acetate. Kelso *et al.*[332] modified the method of Talvitie and Brewer for direct use with water by using an anion-exchange resin in the chloride–acetate forms. Brownley and Howle[111] used a cation-exchange resin to remove undesirable cations from water before direct estimation.

The Intersociety Committee on Methods for Ambient Air Sampling and Analysis has given tentative approval of an ion-exchange method for collected air samples.[305]

The ion-exchange methods are useful for concentrating fluoride ion from large volumes and for separating fluoride ion from other anions and cations. However, because the diffusion method for fluoride is so simple and the distillation procedures are of known reliability, it appears that ion exchange will have only specialized use as a separation scheme.

Distillation

Since Willard and Winter[724] added steam to the Offermann distillation method,[495] this method has been the most important technique for fluoride separation. It is still the standard against which other methods are compared. Over the years, many modifications have been published. Many involved different distillation temperatures and additives to reduce the carryover of contaminants that would interfere with the estimation step;[44,143,178,199,294,492,493,535,603,613,616] a number of these modifications were reviewed by Megregian and Solet.[449] Other modifications have involved the use of multiple still units,[104,415,539,607] jacketed stills,[114,415] and microstills.[596,696]

Fox and Jackson[208] and Fox and Hill[207] have studied the phase relations in the distillation procedure. The studies were directed toward distillation from phosphatic and aluminiferous ores but are applicable to all samples. Their results indicate that the actual form of the fluorine carried over is most likely the complex $H_4SiF_8 \cdot 12H_2O$, rather than fluorosilicic acid, as initially postulated.

During the last few years, a semiautomated microdistillation scheme[316,710,711] was introduced by workers at the Boyce Thompson Institute, and the system has been successfully applied to analysis of air, water, and biologic samples. Derner[176] has applied the system to the analysis of urine.

Warf *et al.*[703] described pyrohydrolysis and applied it to some of the rather specialized needs of the Manhattan project. Newman[477] applied it to aluminum–silicate minerals.

Tentative methods for manual and semiautomated distillation of samples have been published by the Intersociety Committee on Methods for Ambient Air Sampling and Analysis.[305,306]

Diffusion

In 1954, Singer and Armstrong[598] introduced the diffusion method for the separation of fluoride. The method might be considered as a modification of the etch test. It involves the volatilization of fluoride (presumably hydrogen fluoride) in one section of a closed system and absorption of the fluoride in alkali in another section.

In the original method,[598] the authors used a polyethylene bottle as the basic container, with a specially machined strip of polyethylene (on which alkali could be held) as the receiver. The sample, perchloric acid, and the receiver block were placed in the bottle. The bottle was sealed and then tipped or turned to mix the acid with the sample. The entire system was placed in an oven at 50 C for 20 hr; then the receiver was removed and the alkali fluoride rinsed off for estimation. The original method used thorium titration with alizarin as the indicator.

Hall[248,251] suspended filter paper treated with alkali from the bottle cap as the receiver and diffused for 24 hr at 60 C. Baumler[55] and Baumler and Glinz[57] published a similar method. Frere,[219] apparently unaware of the work of Singer and Armstrong[598] and of Hall,[251] developed a diffusion method using a fluoride-resistant plastic Conway dish developed by Öbrink.[490] He acidified with sulfuric acid and diffused at room temperature for an unspecified period. Singer and Armstrong[599] have given more details of a method using the Öbrink cell. Rowley and Farrah[559] used standard 48-mm plastic Petri dishes with alkali spread on the inside of the cover. Chang and Thompson[134] developed a cell that would accept the platinum crucible they used for ashing samples; alkali was spread on a special cover. Nicholson[484] described a multicell disposable-tray system.

The microdiffusion method of separation has had considerable application to the analysis of urine and other biologic materials. In addition to the specific modifications noted above, Debiard and Dupraz[169] applied the method to plant materials and Hall[252] and Cumpston and Dinman[162] to urine. Farrah[201] presented a good discussion of the applicability of the method to routine urine screening of industrial workers. There has been no truly complete study of the factors affecting the method. Wharton[721] reported a partial study of the effect of time and temperature. Hall[249] and Farrah[201] both pointed out that the method provides for separation only of "acid-labile fluoride," and Taves[662] discussed this point in more detail.

The Intersociety Committee on Methods for Ambient Air Sampling and Analysis has given tentative approval to a method in which diffusion is carried out in a disposable plastic Petri dish or in a Conway microdiffusion dish with the Öbrink modification.[305]

CHEMICAL ANALYSIS OF FLUORIDES

Many methods and modifications of methods are available for fluoride analysis. The method selected should be compatible with the earlier collection, separation, etc., and should be relatively simple, accurate, and precise. For analysis of plant tissues, titrimetric and colorimetric methods have been most commonly used, but the fluoride ion selective electrode is coming into wider use. Several methods to be used with both manual and semiautomated procedures have been tentatively approved by the Intersociety Committee on Methods for Ambient Air Sampling and Analysis.[305,306]

Gravimetric Methods

Except for the analysis of fluoride in phosphate rock and other minerals with a high fluorine content, gravimetric methods are not sufficiently sensitive for general use. Only two precipitates have had any wide use—calcium fluoride and lead chlorofluoride (PbClF). McKenna[440] discussed many of the problems of both precipitates and described several nongravimetric methods for estimating the lead chlorofluoride precipitate. Fennell[203] published a method for milligram quantities of fluoride based on precipitation as lead chlorofluorides followed by resolution and titration of the chloride. The reports of Specht[619] and Ergen and Heath[192] provide the best details of the gravimetric procedure.

Enzymatic Methods

The inhibition of some enzymes by fluorides offers an indirect method for estimating the fluoride content of samples. Stetter[629] developed a method based on the inactivation of potato phosphatase and claimed a sensitivity to within about 0.02 μg/ml. Linde[373] described a method based on inactivation of liver esterase and reported that it was suitable for estimating microgram quantities of fluoride. McGaughey and Stowell[438,439] modified Linde's method and reported that they were able to determine fluoride in the range of 5-30 nanograms in a sample volume of 2 ml. Ethylbutyrate was used as the substrate, and the freed butyric acid was titrated with sodium hydroxide.

Titrimetric Methods

Most titration methods are modifications of the original method of Willard and Winter,[724] in which a solution containing fluoride ion is titrated with a standard solution containing an ion that forms a complex with fluoride. The usual complexing agent is thorium. A dye, often alizarin red, is used as an indicator; it forms a colored lake with free thorium ion.

McClure[419] reviewed the modifications of the basic method through 1939. McKenna[440] reviewed the modifications, including indicators other than alizarin, up to 1950. Willard and Horton[723] reviewed in detail the other indicators used for thorium titration. The chemistry involved in the method was elucidated by Horton[295] and by Hodge and Smith.[287] Details of the titration techniques as used in specific laboratories are given by Horton *et al.*,[294] Adams and Koppe,[11] Dean *et al.*,[168] and Mavrodineanu *et al.*[416]

At least two titration methods other than the thorium-dye system have been suggested. Osborn[497] recommended the use of a cation-exchange resin (after, or in conjunction with, appropriate isolation techniques) to form hydrogen fluoride or fluoroboric acid. The free acid is then titrated with a base. Brandt and Duswalt[94] proposed a turbimetric titration with either calcium chloride or thorium nitrate for milligram quantities of fluoride.

Colorimetric Methods

Sensitive color reactions for fluoride ion have been known since around 1908,[452,626] but they had little use. The many color reactions that were suggested have been reviewed by McKenna.[440] Willard and Winter [724] tried to develop a colorimetric method based on deBoer's[170] reaction, but without success. This failure and the success of the titration method may have hindered the development of colorimetric methods for the next 10 years.

In 1943, Talvitie[656] successfully applied the decolorization of the thorium–alizarin sulfonate lake by fluoride; and in 1945, Lamar[348] developed a method based on zirconium–alizarin sulfonate for water analysis. Megregian and Maier[448] and Bumsted and Wells[115] studied many of the substances that interfere and made several improvements in the zirconium–alizarin sulfonate method. Sulfate and phosphate were found to be the major interfering materials.

In 1959, Belcher *et al.*[59] introduced alizarin complexone* (1,2-dihydroxyanthraquinonyl-3-methylamine-*N,N*-diacetic acid), which forms a red chelate with cerium (III) and is converted into a blue complex in the presence of the fluoride ion. Unlike the bleaching of color of the metal–alizarin sulfonate lakes by fluoride, the metal–alizarin complexone chelates form new, stable, colored complexes of metal–fluoride–alizarin complexone. The alizarin complexone chelates are as sensitive as the alizarin sulfonate systems to interference by sulfate and phosphate. Two complexes are in general use: the cerium (III)–alizarin complexone initially used by Belcher

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*Also known as "alizarine complexone," "alizarin complexon," "alizarin complexan," and "alizarin fluorine blue."

et al.[59] and applied by Frere,[210] Yamamura *et al.*,[732] and Belcher and West;[60] and the lanthanum (III)–alizarin complexone suggested by Frere[210] and applied by Hall,[252] Wade and Yamamura,[696] Cox and Backer,[156] and others. Ross *et al.*[558] used the cerium (III)–alizarin complexone in their semiquantitative field spot test for fluoride in vegetation, and Weinstein *et al.*[710,711] used the lanthanum (III) form in their semiautomated procedure. Yamamura *et al.*[732] presented the most complete description of the method based on cerium with a reasonably complete study of interfering ions. The method based on lanthanum was described in detail by Wade and Yamamura[696] and Sultzburger.[23] The lanthanum system may be slightly less sensitive than the cerium complex, but it has a somewhat greater range and is less sensitive to interference.[23]

Most of the dyes used as indicators for the thorium titration[723] and several others[440] have been suggested for use in color reactions for estimating the concentration of the fluoride ion. Revinson and Harley[538] proposed the thorium–Chrome Azurol S system; Powell and Saylor[527] and MacNulty and co-workers[74,300,400,401,403] studied several aluminum–dye systems; and Nichols and Condo[483] examined 18 iron (III) complexes. In 1954, Megregian[447] applied the zirconium-Eriochrome Cyanine R complex to water analysis. This reaction, like the zirconium–alizarin system, depends on the bleaching action of fluoride ion on the metal–dye complex.

The zirconium–Eriochrome Cyanine R complex became the method of choice for many workers.[162,169,485,486,492,493,559,596] It proved to be almost as sensitive as the zirconium–alizarin system and required a shorter period for color development. Adams and Koppe[10] used this reagent in their automatic air-analysis instrument. Megregian,[447] Oelschläger and Kirchgessner,[493] and Oelschläger[492] provide complete descriptions of the method and of its limitations.

In 1958, Bellack and Schouboe[63] introduced the dye, sodium 2-(*p*-sulfophenylazo)-1,8-dihydroxynaphthalene (SPADNS), in a zirconium–dye system for water analysis and suggested the reagent as a substitute for the zirconium–alizarin complex. It is less sensitive than the zirconium–alizarin complex, but it is stable, has a broader absorption spectrum, and reacts more rapidly with fluoride ion. This permits the use of simple filter photometers for measurement. The system has had considerable use[134,332,484,657] and can be easily substituted in any method based on the zirconium–alizarin system.

Other color methods have had limited use. Bartlet *et al.*[54] used the bleaching effect of fluoride on the ferric salicylate color, and

Brownley and Howle[111] used the thorium–phenylfluorone system. Adams[3] has used the iron–ferron complex in an automatic air sampler for high atmospheric fluorine concentrations. Ivie *et al.*[308] used the quench of the fluorescence of magnesium-8-hydroxyquinoline by hydrogen fluoride as the basis for an automatic air-analysis instrument. Bumsted and Wells[115] suggested strontium chloranilate, and Hensley and Barney[271] suggested thorium chloranilate. Ballczo[52] used $LaTl[Fe(CN)_6]$ in a sensitive semiquantitative spot ring test. Weisz[717] developed a spot test based on the release of rhodizonate from iron (III) rhodizonate by fluoride. The free rhodizonate reacts with silver ion to form a blue-black complex. Brandt[91] in his brief review noted several other color reactions of potential use.

The most common and most adaptable color methods in use at present are the zirconium–alizarin, the zirconium–SPADNS, the zirconium–Eriochrome Cyanine R, and the lanthanum–alizarin or cerium–alizarin complexone systems. There are some differences among these reagents in their sensitivity to interference from other materials, but with only minor modifications in overall procedures, the reagents are almost all interchangeable.

Electrochemical Methods

MacNulty *et al.*[402] developed an indirect polarographic method for fluoride using aluminum, and Shoemaker[584] developed a similar procedure based on iron. The latter method was developed for use on radioactive solutions so "hot" that the usual metal–dye systems were bleached by the radiation. The author reported that he could estimate 1 μg of fluoride to within 15%.

Dubois *et al.*[181] described a method based on the depolarization of the aluminum–platinum electrode by halogen ions. They used the rotating aluminum electrode system of Kolthoff and Sambucetti[338–341] and were able to suppress the effect of chloride and other halogen ions by careful bias control. When applied to urine, the method compared well with standard color procedures.

In 1966, Frant and Ross[209] announced the development of a fluoride ion selective electrode. Operation of the electrode is similar to the familiar glass electrode used to measure hydrogen ion activity. The electrode does not measure fluoride ion concentration, but rather ionic activity. Provided that various other characteristics in the solution are controlled, activity is easily translated into concentration. Lingane[374] has published the preliminary operating factors for the electrode and has applied the electrode to determining the end point of the titration of moderate amounts of fluoride ion (50–80 mg) with thorium, lanthanum, and calcium. Durst and Taylor[183] have modified the electrode to handle volumes as small as 50 μl. Babcock and Johnson[50] and Andelman[31] reported on the use of the electrode in a water-supply-monitoring system. Crosby *et al.*,[159] in their review of methods of fluoride ion analysis for waters and aqueous solutions, gave comparisons of the fluoride ion electrode with standard colorimetric methods. The fluoride ion electrode was compared with an ion-exchange method using zirconium–SPADNS for color development in a collaborative study of water analysis among 104 laboratories.[436] Satisfactory results were obtained by both methods, but the simplicity of operation of the electrode made it the method of choice. Elfers and Decker[187] and Chapman[135] have used the electrode in the analysis of air and stack-gas samples and obtained results comparable with those of the zirconium–SPADNS method. Methods have also been published for the analysis of fluoride in blood plasma, urine, bones, and teeth,[53,597] feeds,[678] and phosphate.[384] Jacobson and Heller[314] recently reported a method for the direct analysis of fluoride in plant tissues without prior ashing, alkali fusion, or separation. Undoubtedly, as experience is gained and the limits are better defined, the fluoride ion selective electrode will find increasing use.

SUMMARY

Manual methods for sampling air for fluoride are only reasonably adequate for the needs. Methods for sampling total fluoride have been reviewed.[202,505–507] Techniques for separating particulate and gaseous fluorides are available,[202,244,407,507,674] but the separation is not perfect and a standard reference method is not available. One automatic instrument is available,[9,10] and at least two others are in development.[579,667,674] A sensitive, continuous, dependable instrument with a short averaging time is needed. The lime–filter-paper method may be useful for determining the relative distribution and amounts of atmospheric fluoride.[4,5,458,547,726]

There are many methods of fluoride analysis adequate for almost all needs. Any of several equally good estimation procedures are available, with the choice of method depending only slightly on the amount of fluoride to be measured, and more on its simplicity and the personal preferences of the analyst.

Tentative methods of sample preparation; separation by ion exchange, distillation, and diffusion; and determination by titrimetric and colorimetric methods have been published by the Intersociety Committee on Methods for Ambient Air Sampling and Analysis.[305,306]

5

Is Fluorine an Essential Element?

The generally accepted criteria that determine the essentiality of any element have been well documented by Underwood.[685] First, it should be possible to demonstrate repeated and significant responses in growth or health to dietary supplements of the element and to the element alone; second, it should be possible to develop a deficiency state on diets that lack the element but that are otherwise adequate and satisfactory. Such diets should contain all other known dietary essentials in adequate amounts and should be free from toxic compounds. With most elements that have been shown to be essential, it has also been possible to show definite correlation between amounts of the element present in various body organs and amounts present in the diet.

Unequivocal evidence that fluorides perform any vital function in animals has not yet been produced.

There is no doubt that an increase in the fluoride intake of the population in most parts of the world does result in a decrease in incidence of dental caries in that population. That in itself is no indication of fluorine essentiality, inasmuch as caries incidence depends on many factors, and many persons with perfectly sound dentition have had only minimal exposure to fluoride. The observation of Bernstein *et al.*[70] that increased intake of fluoride may be of value in the prevention of various osteoporotic diseases falls into much the same category.

Attempts of early nutritionists to develop a diet that was sufficiently low in fluoride to determine whether fluorine was an essential element were hampered by the widespread occurrence of fluorides in their dietary ingredients. Most of the early studies (see Muhler[468]) were carried out with diets that contained at least 0.1 ppm of fluoride, and perhaps even more. More recently, two investigators have produced diets that are extremely low in fluoride content and have failed to find any indication that fluorine is an essential element. Maurer and Day,[413] by carefully purifying all dietary ingredients, produced a diet that contained about 0.007 ppm of fluoride and raised rats on it through four generations. When the performance of these animals was compared with that of animals that were fed the same diet but given 2 ppm of fluoride in their drinking water, it was found that there was no difference in general health, dental health, weight gain, or phosphatase activity in various tissues. In an attempt to develop a diet with even less fluoride, Doberenz *et al.*[179] developed a diet based on the use of hydroponically grown soybeans and sorghum. They formulated a diet that apparently contained less than 0.005 ppm of fluoride. There was no detectable difference in the general health or growth rate between the rats fed that diet and the rats fed the same diet plus 2 ppm of fluoride in their drinking water. A large number of serum and tissue enzyme activities were determined, and the only significant difference was a slight increase in the activity of serum isocitric dehydrogenase in rats on the low-fluoride diet. It was concluded from this study,[179] as from the Maurer and Day study, that there was no reason to consider fluoride as a dietary essential for the rat. A similar study from the same laboratory[706] failed to find any indication that fluoride was essential for mice raised through three generations on an extremely low-fluoride diet.

It cannot definitely be concluded from these experiments that fluoride is nonessential for the nutrition of these species. Different results might be obtained if it were possible to produce diets still lower in fluoride. It is nevertheless apparent that, if fluoride is a dietary essential for the species studied, its requirement must be extraordinarily low. Indications that fluoride might be essential for any other species are also lacking, and the beneficial effects of fluoride on dental health or bone metabolism should be considered as pharmacologic responses, and not as a cure of pre-existing deficiency condition.

A number of studies have been done over the last 100 years to determine whether fluorine is essential for plant growth.[67] [71] None of these studies was convincing. The only credible (but as yet unsupported) evidence available to date is the recent report of Bertrand,[73] who concluded that fluorine is essential for corn growth and development when aluminum is present in the growth medium.

6

General Aspects of the Effects of Fluoride on Living Systems

The effects of fluoride on living systems have been considered at various levels of cellular and tissue organization. The response of isolated enzymes to increased concentration of fluoride has been determined, and a wide range of sensitivities have been observed. A considerable amount of work has been done to ascertain the effects of adding fluoride to the incubation media of intact cellular preparations, including cultured mammalian cell lines, erythrocytes, liver slices, bone slices and cultures, algae, and plant leaf, root, and coleoptile preparations.

The biochemical effects of fluoride on separate cells and tissues may be very different from the effects on intact plants and animals. Plasma fluoride concentrations of animals that have ingested chronically toxic amounts of fluoride have seldom been observed to exceed 0.1 m*M*, and plasma fluoride concentrations of animals that succumbed to acutely toxic doses of fluoride were about 1.0 m*M*. Most of the *in vitro* effects of fluoride occur at concentrations considerably above this. The fluoride content of plant tissue exposed to gaseous fluorides does increase drastically, but little is known about the distribution or availability of fluoride within the cells.

EFFECTS OF FLUORIDE ON ENZYMES

The effects of fluoride on enzyme activity have been reviewed on several occasions since Borei's[85] extensive review. Thomas and Alther[671] have considered the effects of fluoride on some enzymes involved in plant metabolism, and Matthews[411] has recently discussed changes in cell function that may be mediated by the effects of fluoride on enzymes. Wiseman[727] has extensively reviewed the nature of the *in vitro* inhibition of purified enzyme preparations and has provided an excellent review of the current knowledge in this area. Venkateswarlu[694] has also summarized several examples of fluoride activation and inhibition of purified enzymes.

Fluoride inhibits many enzymes and undoubtedly does so through different mechanisms. Enolase is subject to inhibition due to the formation of a fluoride–phosphate–magnesium complex. Magnesium-activated phosphatase and pyrophosphatase, manganese- or magnesium-activated phosphoglucomutase, and iron-containing succinic dehydrogenase are fluoride-sensitive and may be inhibited by the formation of a metal–fluoride–phosphate complex. Fluoride inhibits peroxidase, catalase, and cytochrome oxidase by combining with ferric iron in their heme groups. Fluoride inhibits some enzymes that do not have a metal ion requirement and must do so by direct interaction with the enzyme or substrate. It should be considered that, in addition to having a relatively specific effect on particular enzymes, fluoride may exert a general effect on metabolism by its formation of metal–fluoride complexes. The altered nutrient status of the cell could result in decreased activity of its metal-requiring enzymes, or, as Hewitt[273] has suggested, it could cause a weakening of its structural integrity if calcium were involved.

There has been a tendency to emphasize the role of fluoride as an inhibitor of glycolysis by virtue of its inhibition of enolase. However, through its inhibition of other enzymes, it inhibits energy metabolism via the tricarboxylic acid cycle both by blocking the entry of pyruvate and fatty acids and by inhibiting succinic dehydrogenase. Fluoride should therefore be thought of not simply as an inhibitor of glycolysis, but rather as a general inhibitor of oxidative metabolism.

The question of which enzymes are most sensitive to fluoride is complicated by the fact that sensitivity is influenced by many other factors. On the basis of his review of *in vitro* studies with various enzyme preparations, Wiseman[727] has listed enzymes that are most sensitive at pH values that might be encountered *in vivo*. They are shown in Table 6-1.

TABLE 6-1 Enzymes Highly Sensitive to Fluoride in pH Range 5–8[a]

Enzyme	Molar concentration of fluoride[b]	Inhibition, %
Human salivary acid phosphatase	2×10^{-4}	55
Erythrocyte inorganic pyrophosphatase	2×10^{-5}	52
Human plasma cholinesterase	5×10^{-5}	61
Sheep brain glutamine synthetase	5×10^{-5}	50
Liver methionine "activating" enzyme	8×10^{-4}	28
E. coli metaphosphate synthetase	5×10^{-4}	100
Pterin deaminase (bacterial)	3×10^{-5}	50
Glucose 1-phosphate dismutase (*E. coli*)	5.8×10^{-4}	75

[a] Modified from Wiseman.[727]
[b] 1 ppm = 5.25×10^{-5} *M*.

Capozzi *et al.*[122] have recently investigated the effects of six inorganic fluoride compounds on nine glycolytic enzymes *in vitro*. The study is noteworthy because each fluoride was tested at the same two concentrations (38 and 76 ppm, or 2 and 4 m*M*, respectively), permitting valid comparisons of the sensitivities of the enzymes. The fluoride concentrations used, however, are appreciably higher than those encountered *in vivo*. The data are shown in Table 6-2.

Venkateswarlu[694] has listed examples of physiologic processes and enzymes that are influenced slightly to moderately by fluoride concentrations below 2 ppm; these are shown in Table 6-3. Venkateswarlu points out that little is known about the mechanisms whereby fluoride stimulates enzyme activity. He calls attention to the situation in which, when a particular metabolic pathway is blocked by fluoride, an alternative pathway may appear to be or may actually be more heavily used, thus producing an appearance of activation.

EFFECTS OF FLUORIDE ON CELLS AND ISOLATED ORGAN AND TISSUE SYSTEMS

Most studies of the effects of fluoride on mammalian cell or organ cultures have been directed only toward determining the concentrations of fluoride that inhibit cell replication and growth. Berry and Trillwood[71] reported that the growth of HeLa cells and mouse fibroblasts was extremely sensitive to less than 1 ppm of fluoride in the culture media. This extreme sensitivity has not been substantiated by Armstrong *et al.*[45,46] or Nias.[481]

TABLE 6-2 Effects of Selected Fluoride Compounds on Glycolytic Enzymes *in Vitro*[a] Enzyme Activity[b]

Enzyme	Control	Na_2PO_3F		$MgSiF_6$		Na_2SiF_6		NaF		SnF_2		GA 297[c]	
		2 m*M*	4 m*M*	2 m*M*	4 m*M*	2 m*M*	4 m*M*	2 m*M*	4 m*M*	2 m*M*	4 m*M*	2 m*M*	4 m*M*
HK	95.8	95.8	107.5	93.5 (2.4)	90.8 (5.2)	74.4 (22.4)	78.9 (17.6)	90.8 (5.2)	90.8 (5.2)	90.8 (5.2)	–	88.5 (7.7)	98.1 (2.4)
F-6-PK	68.5	68.5	56.8 (17.1)	55.2 (19.4)	43.0 (37.2)	43.0 (37.2)	30.7 (55.2)	68.5	53.7 (21.7)	–	–	62.2 (9.2)	55.2 (19.4)
GAPD	479	450 (6.1)	364 (24)	364 (24)	258 (46.2)	248 (48.2)	240 (50.0)	431 (10.2)	325 (32.2)	364 (24)	–	287 (40.2)	325 (32.2)
PGM	68.2	64.9 (4.8)	38.6 (43.3)	38.3 (43.7)	16.9 (75.2)	16.9 (75.2)	10.3 (84.5)	19.2 (71.8)	9.6 (85.5)	31.3 (54.2)	16.9 (75.1)	27.5 (59.5)	8.4 (87.5)
PGK	27.5	28.7	25.2 (8.5)	20.5 (25.5)	20.5 (25.5)	20.5 (25.5)	5.5 (80.0)	21.6 (21.4)	22.2 (20.1)	22.0 (20.1)	–	26.4 (4.1)	26.8 (2.5)
PK	163	159 (2.4)	125 (24.4)	115 (29.5)	101 (38.1)	144 (11.6)	101 (38.1)	115 (29.5)	104 (36.3)	115 (29.5)	–	144 (11.6)	125 (24.4)
ENOL	36.1	27.6 (23.6)	17.2 (52.2)	10.9 (69.0)	7.1 (80.1)	6.0 (83.2)	2.0 (92.0)	6.0 (83.2)	2.9 (92.0)	18.7 (48.2)	14.4 (60.0)	9.5 (73.5)	3.7 (89.5)
LD	358	344 (3.9)	287 (19.8)	278 (22.4)	364	239 (33.2)	258 (28.0)	192 (46.8)	139 (33.2)	210 (41.4)	249 (30.5)	325 (9.2)	383
G-6-PD	123	123	123	116 (5.7)	123	120 (2.4)	123	100 (18.7)	123	116 (5.7)	123	123	135

[a] Reprinted with permission from Capozzi *et al.*[122]
[b] Enzyme activity is expressed as international units per milligram of protein. Values in parentheses are percent inhibition induced by fluoride compounds. Molarity indicates concentration in reaction mixture.
[c] *N,N',N'*-tripolyoxyethylene-*N*-octadecylpropylenediamine dihydrofluoride.

TABLE 6-3 Physiologic Processes and Enzymes Influenced Slightly or Moderately by Low Fluoride Concentrations (10^{-4} *M*/liter and below)[a]

Physiologic Process or Enzyme Involved	Source or Material	Fluoride Concentration, *M*/liter[b]
	Activation by Fluoride	
Glucose fermentation	*E. coli*	10^{-5}
Embryonal culture	Chick heart	1.25×10^{-5}
Photosynthesis	Kidney bean (*Phaseolus vulgaris*)	3×10^{-5}
Fermentation	Yeast	5×10^{-5}
Respiration	Cancer tissue	10^{-4}
Nitrogen fixation	*Azotobacter*	10^{-4}
Lipase	Liver	5×10^{-5}
	Inhibition by Fluoride	
Calcification	Hypertrophic cartilage slices (rat)	10^{-5}
Citric acid production	*Aspergillus niger*	5×10^{-5}
Acid production	Streptococci and lactobacilli	10^{-5}
Activation of acetate	Kidney and liver homogenates	10^{-4}
Esterase	Pig liver	5×10^{-7}
Acid glycerophosphatase	Sheep brain	6×10^{-6}
Carbonic anhydrase	Rat incisor	10^{-5}
Adenosine triphosphatase	Cardiac muscle	10^{-5}
Phosphomonesterase	Serum	10^{-5}
Phosphatase	Coconut kernel	10^{-4}
Pyrophosphatase	Yeast	10^{-4}
Pyrophosphatase	Firefly	10^{-4}
Glutamine synthetase	Animals and plants	10^{-4}
Acid phosphatase	Human saliva and prostate gland	10^{-4}
Isocitric dehydrogenase	Rat liver	10^{-4}

[a] Adapted from WHO.[731] (p. 183)

[b] The fluoride concentration in normal body fluids and soft tissues is 5×10^{-6} to 5×10^{-5} *M*/liter. "Ionic" fluoride in plasma is 5×10^{-7} to 2×10^{-6} *M*/liter.

Albright[20] found that the replication of murine leukemic lymphoblasts was unaffected by 5–6 ppm of fluoride but noted a sigmoidal relation between the percentage of cell growth and fluoride concentration. At higher concentrations, a slight inhibitory effect was detected at 8–9 ppm of fluoride, and a rapid depression of growth between 11 and 55 ppm. Similar results were obtained by Carlson and Suttie[125,126] with HeLa cells. The minimal concentration of fluoride required to inhibit growth (measured as accumulation of cell protein) was 20 ppm, whereas 40–45 ppm was necessary to stop growth completely. Pace and Elrod[500] have found a similar range of toxicity for L cells.

Recently, LeCoultre-Mulder *et al.*[357] reported a study in which HeLa, T, and amnion cells were found to be sensitive to as little as 2 ppm of fluoride. The conclusions, however, do not seem to be well supported by the statistical data presented.

The mechanism by which fluoride affects growth in these cellular systems is not known, and little work on possible metabolic alterations has been done. Carlson and Suttie[126] found that limited fluoride inhibition of HeLa cell growth was associated with a decreased efficiency of glucose utilization, which may have been associated with lower growth rate. The percentage conversion of glucose to lactic acid was not altered by fluoride. Short-term experiments with $[^{14}C]$-U-glucose indicated that the $^{14}CO_2$ production was unaffected by growth–inhibiting levels of fluoride. If fluoride was present during the entire experimental period, HeLa cells maintained an adenosine triphosphate (ATP) concentration only slightly below that of control cells; but if fluoride was added to rapidly growing cells, a rapid decrease in cellular ATP was observed.

The effect of fluoride on cells will depend on the intracellular fluoride concentration, which has seldom been determined.

Partridge[511] has found that the ratio of fluoride in intracellular water of HeLa or L cells to that in their media was between 0.3 and 0.4 and that the ratio was unchanged when the media fluoride was varied from 10 to 50 ppm. The ratio in other cell lines is probably very similar.

Studies of oxygen uptake and phosphate incorporation in *Chlorella* indicate that the effect of fluoride may be more complex. Both processes have been shown to be stimulated by fluoride over a wide range of fluoride concentrations. Beyond some concentration, however, an increase in the fluoride concentration resulted in increased incorporation of phosphate into nucleotides but not in increased oxygen uptake.[442]

Matthews[411] has reviewed the effects of fluoride on a large number of different animal tissue preparations. In most of these studies, the use of fluoride was peripheral to the main purpose of the experiment, and the data contribute little to an understanding of the toxic actions of fluorides.

Bone slices have been used to study the effect of fluoride on calcification, and both inhibition[241,509] and enhancement[654] have been observed. The interrelationship between fluoride and parathyroid hormone in bone cultures has been studied by Raisz and Taves[531] and by Goldhaber.[233] It was found that between 2 and 9 ppm of fluoride was needed to decrease the hormone-mediated release of calcium from these bone preparations.

Isolated plant tissues have also been used to study the effects of fluoride on metabolic systems *in vivo* and the effects of various factors on sensitivity to fluoride. These experiments with isolated plant tissues have shown that a variety of metabolic pathways may be affected by fluoride and that the presence or type of effect depends on which pathways are operative.

Pea epicotyl sections,[142] avena coleoptiles,[496] and root tip preparations[522] are some of the experimental plant materials that have been used to study the effects of fluoride.

METABOLIC EFFECTS OF FLUORIDE ON INTACT PLANTS AND ANIMALS

Although it is clear from the information available on fluoride-sensitive enzymes that fluorides are potential metabolic inhibitors, there is little information to indicate what metabolic lesions might be present in the tissues of intact animals whose diets contain high concentrations of fluoride. Even a casual search of the literature reveals that most of the enzymes that are usually considered to be rather sensitive to fluoride do not exhibit appreciable inhibition until the fluoride concentration reaches 10^{-3} *M* or even 10^{-2} *M*. In contrast, the concentration of fluoride in plasma and soft tissues of animals fed large amounts of fluoride remains very low. In the rat, for example, 600 ppm of fluoride in the diet raises the plasma fluoride content to only about 3 ppm (1.5×10^{-4} *M*).[593] Although some enzymes have been reported to be sensitive to fluoride at that concentration, one cannot simply extrapolate from *in vitro* enzyme studies to determine the effect of fluoride on animal metabolism.

The metabolic effects of fluoride ingestion in animals are complicated by the effect of fluoride on food intake[638] and the alterations in metabolism that go with it. A decrease in food intake is to some extent related to plasma fluoride content. Shearer and Suttie[583] have postulated that the relation between food intake and plasma fluoride concentration is indirect and mediated by an effect of fluoride on tissue enzyme activity that results in a shift in the concentration of some metabolite(s). The altered balance of tissue metabolites, rather than the increase in fluoride concentration, is thought to be the signal that reduces dietary intake.

In an attempt to locate points of fluoride inhibition in carbohydrate metabolism, Shearer and Suttie[582] determined the liver concentrations of many glycolytic and some Krebs cycle intermediates in rats fed 450–600 ppm of fluoride (as sodium fluoride) for 3 days. Fluoride ingestion caused a small decrease in a number of metabolites, rather large decreases in pyruvate and lactate, and a large increase in liver citrate. With the exception of the increase in liver citrate, the changes appeared to be secondary to the direct effect on food intake. That would also appear to be true for the changes in carbohydrate metabolism[739] and the liver enzyme changes[127] previously observed. The latter study[127] seems to be the first to demonstrate an *in vivo* inhibition of hepatic enolase in the rat after intraperitoneal injection of fluroide. The ingestion of 600 ppm of fluoride in the diet, however, did not affect hepatic enolase activity.

The key metabolic lesions present in the tissues of animals fed toxic amounts of fluoride remain to be determined. Research activity in this direction has been very limited, but with the new techniques available, knowledge in this area should increase rapidly.

In contrast with animal soft tissues, metabolically active plant cells do accumulate high concentrations of fluoride. Gaseous fluorides are readily absorbed by leaves through the stomata and possibly through the cuticle as well. If adequate moisture is present, deposits of fluoride salts are absorbed by leaves, probably in proportion to their solubility. Once fluoride has penetrated a leaf, it is rapidly transported in the transpirational stream[317,730] and accumulates at the leaf tip and margins, but there is little or no further movement from leaf to leaf or from the leaf to stems, roots, or other organs.[358]

The manner in which atmospheric fluorides affect the metabolism of plants is not well understood. That is due to the limited knowledge of the normal metabolic processes of plants, the manner in which they are integrated and controlled, and the metabolic effects of fluoride itself. The biochemical effects of atmospheric fluoride involve many processes that occur at different times and on different levels of organization. Therefore, any investigation of those effects must use various experimental systems.

Several investigators have measured enzyme activities or pool sizes of metabolic intermediates in fumigated plants and such physiologic processes as photosynthesis and respiration in plants exposed to atmospheric fluorides. Specific examples are described in Chapter 7.

7

Effects of Fluoride on Vegetation

Plants are receptors of fluoride from three sources in their immediate environment: soil, water, and air. There is really no distinction for a plant between soil and water as sources of fluoride, in that both contribute fluoride to the soil solution, the microenvironment of the roots. The phytotoxicity of fluoride from the soil or water is relatively unimportant, although the form and amounts of fluoride in or on a plant may be consequential for foraging animals.

Phytotoxicity of fluoride usually is attributable to airborne forms—e.g., hydrogen fluoride, silicon tetrafluoride, and such soluble particulate materials as sodium fluoride and aluminum fluoride. Other forms of fluoride are less phytotoxic but are of concern when accumulated by plants and transmitted to foraging animals.

Once fluoride has entered a plant, it moves in the transpirational stream. Thus, fluoride taken up by the roots passes from them and through the stem, to accumulate in the foliar tissues and ultimately in the apical or marginal portions of leaves. Fluoride from the atmosphere is preponderantly received by foliar tissue and moves to the marginal or apical portions of leaves. The fluoride in a leaf is generally immobile; it does not move to other leaves or organs of the plant.

The accumulation of fluoride in the foliar tissue can be accompanied by observable effects, depending on the relative susceptibility of the plant; the environmental and biologic factors operating before, during or after exposure to fluoride; the atmospheric concentration of fluoride; and the duration and frequency of exposure.

In this chapter, the greatest reliance will be placed on reports that relate effects of fluoride to known times and concentrations. Thus, data obtained by spraying plants with solutions of hydrogen fluoride or fluoride salts will be considered only if they clarify or help to explain the plants' response to airborne fluoride.

A considerable portion of this chapter is based on the excellent review, "Report on Fluorides," prepared by the U.S. Department of Agriculture for the National Air Pollution Control Administration.[92]

Any plant or population of plants may serve three different functions: commercial, aesthetic, or ecologic. The commercial functions are exemplified by agriculture and forestry, in which the plants are the basis for the production of food, forage, fiber, and fuel. The aesthetic function of plants is most evident in private gardens and public parks. The ecologic function is very complex and includes oxygen production, provision of habitats and food for wildlife, and roles in soil and water conservation. Of course, any plant or lot of vegetation can serve all these functions simultaneously, and particular plants can be grown on the basis of their suitability to a desired function. The effect of atmospheric fluoride on a plant becomes significant when it is related to the function of the plant.* However, most of what is known about the effects on plants has been presented as the product of a scientific investigation and has not been presented in an immediate or practical context. Consequently, many effects described here must be interpreted with respect to the functional role of plants in order to evaluate their significance.

THE NATURE OF EFFECTS

One concise way to summarize the response of vegetation to fluoride is by a scheme after that used by Vogl *et al.*[695] for the effects of sulfur dioxide and modified by Weinstein and McCune for fluoride[713] (Table 7-1). In this scheme, one considers effects classified by the level of biologic organization and the degree and type of effect produced at each level. Thus, an attempt is made to relate the effects at the cellular level with those anticipated at the level of the intact plant or plant community.

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*The concentration of atmospheric fluoride has been expressed in different ways by investigators over the years. As noted in Chapter 1, effects on vegetation are discussed in terms of micrograms of fluoride per cubic meter ($\mu g/m^3$); a concentration of fluoride (as hydrogen fluoride) of 1 part per billion (ppb), by volume, is equivalent to 0.8 $\mu g/m^3$.

For example, the well-known inhibition by the fluoride ion of many enzymes *in vitro*, requiring divalent cations, is postulated as one type of metabolic effect *in vivo*. If the enzyme constitutes a rate-limiting step, either naturally or through the effects of fluoride ion, in a pathway consisting of several enzymes, one can expect that an inhibition of the entire pathway will occur.

There is evidence that alternate pathways are used in glucose catabolism when plants are exposed to fluoride.[382,427,556,557] Thus, respiratory activity may be maintained, but the production of secondary metabolites may be altered because the two pathways do not yield the same intermediates. If enough metabolic sites are affected or a major pathway is sufficiently inhibited, modification, disruption, or death of the cell could occur.

TABLE 7-1 Nature of Fluoride-Induced Effects in Plants at Four Levels of Biologic Organization

Cellular	Tissue or Organ	Organism	Ecosystem
(Altered cell milieu?)			Increased F in ecosystem
Effects on enzymes and metabolites			
	Decreased assimilation		
	Altered respiration		
Modification of cell organelles and metabolism	Altered growth and development	Modified growth	Increase in F burden of animals
Pathway disruption			
	Chlorotic lesions	Reduced reproduction	Fluorosis in animals
Cellular modification			
Disruption and death of cell	Necrotic lesions	Decreased fitness for environment	Change in plant community
	Death or abscission of leaf	Death of plant	
			Desolation

The metabolic effects of fluoride at the cellular level would be imparted to the next higher level of organization. For example, one can postulate that the altered patterns of oxygen uptake or carbon dioxide assimilation observed in tissues or organs reflect altered metabolism or modifications in cell organelles. The obvious result on the organ level of cellular modification or disruption is the occurrence of foliar lesions, the best-documented response of plants to fluoride. There is considerable variation in the foliar lesions, depending on the class of plants (narrow-leaved or broad-leaved), their relative susceptibility, and the concentration and duration of exposure.

When the assimilatory capabilities of a single leaf are reduced through abscission, necrosis, chlorosis, or reduced surface, and that leaf is representative of the foliar mass of the plant, then one might logically translate an effect from the organ level to the entire plant. Such effects at the level of the organism might be manifested by modified growth, reduced reproduction, decreased fitness for the environment, or even death. The effects that involve individual plants may in the aggregate influence the ecosystem. The more obvious effects would be registered as changes in the composition or structure of the plant community, with consequent effects on fauna. With fluoride, however, changes in the ecosystem can be brought about, not as a direct result of effects on plants, but through transfer of fluoride from plants through the ecosystem. Thus, a thread of inference connects degrees of effects at each level of biologic organization.

Metabolic Responses

The metabolic effects of fluoride involve many processes that occur at different times and at different levels of organization. Therefore, any investigation of them must use different types of experimental systems. Studies of enzyme activity *in vitro* have provided most of the information on the nature of fluoride-sensitive sites and the possible mechanism of fluoride inhibition. Isolated tissues have been used to study the effects of fluoride on metabolic systems *in vivo* and the effects of various factors on fluoride sensitivity. Entire plants fumigated with hydrogen fluoride have been used to study the biochemical effects of atmospheric fluoride and their relationship to growth, yield, and injury. It is unfortunate that no single system could yield a complete answer, because the extrapolation of the results of one system to another involves many assumptions of uncertain validity.

ENZYMES AND METABOLITES

Because fluoride salts have long been standard inhibitor reagents on the shelf of the enzymologist, it is natural that the effect of fluoride on important plant, animal, and microbial enzyme systems has received much attention.[274] Several workers have presented data on the effect of fluoride on levels of enzyme activity in plant tissue.[133,359,376,377,433,655] Tager and Rautanen[655] found no effect on the sulfite-to-sulfate oxidation systems. Others[359,433] reported stimulation, no effect, or reduction in activity of a variety of enzymes. Lovelace and Miller[377] found that fluoride inhibited succinate oxidation *in vitro* and apparently confirmed it *in vivo*.[376]

The concentrations or pool sizes of various metabolites are also affected by fluoride without attendant visible changes in the plant. Changes in keto acids,[433] organic acids, amino acids and amides, free sugars, DNA and RNA phosphorus,[709] and starch and nonstarch polysaccharides[7] have been found in plants fumigated with hydrogen fluoride. Neither Pack and Wilson[508] nor McCune *et al.*[434] found changes in phosphate esters of plants exposed to hydrogen fluoride, although the incorporation of phosphorus-32 was decreased after corn foliage exhibited mild chlorosis.[434]

CELL ORGANELLES

Several studies[444,479,480,523] have confirmed that fluoride reduces the chlorophyll content of plant tissue. The mechanism and significance of that fact are not clear,[479] but the presence of chlorosis, a visible symptom of fluoride injury in some species, may be regarded as a manifestation of altered structure and therefore function of chloroplasts.

Mohamed and his co-workers[461-464] reported on the development of chromosomal aberrations in tomato, corn, and onion roots, as a result of fumigation with hydrogen fluoride or treatment with sodium fluoride solutions. An increase in phenotypic abnormalities in the second generation of tomato was described. The work has not yet been confirmed by other investigators.

METABOLIC PATHWAYS

Perhaps the best-documented effects of fluoride on a metabolic pathway are found in carbohydrate metabolism. Christiansen *et al.*,[141] Laties,[356] and Bonner and Wildman,[84] through the use of inhibitors and exogenously supplied substrates, have demonstrated the inhibition of glucose catabolism in isolated plant tissues. ^{14}C-labeled glucose has been used frequently to estimate the relative contributions of various pathways to overall glucose catabolism. Ross *et al.*[556,557] have suggested that exposure to fluoride reduces the activity of the glycolytic pathway relative to the pentose phosphate pathway. But Lustinec *et al.*[382] showed that the apparent fluoride sensitivity of the glycolytic pathway is highly dependent on the age of the tissue. McCune *et al.*[427] also reported that fluoride alters the pathways of glucose catabolism and suggested that these effects represented modification of the normal changes that accompany growth and development of the leaf.

CELLULAR MODIFICATION

Histologic changes are associated with the presence of injured tissue, although their spatial relation may vary in different plant species. Solberg and Adams[617] reported that the cellular manifestations were characterized by collapse of the mesophyll and concurrent distortion and collapse of epidermal cells in broad-leaved plants. Cellular collapse was noted in pine needles,[618] but there was considerable variation from tissue to tissue within a needle. Perhaps the most interesting effect was hypertrophy of epithelial cells of the resin duct. In geranium leaves, Poovaiah and Wiebe[525] reported distortion of chloroplasts and starch grains in cells near the injured area, whereas tyloses were induced in the vessel elements of the petiole, an effect also associated with senescence of the leaf.

There is no clear demarcation in metabolic effects between the cellular and the organ levels, inasmuch as they are defined only by the nature of the experimental object. Therefore, studies on oxygen uptake or carbon dioxide assimilation by leaves, while reflecting the aggregate of cellular activities, are the logical extension of studies of effects at the cellular level. An alteration of oxygen uptake or carbon dioxide evolution by plants exposed to fluoride is a result of changes in glucose catabolism in the respiratory pathways of the cell and is therefore of secondary importance at the organ level. Nevertheless, effects on this process have been studied extensively.[40,42,43,276,381,441-443,445,709,735]

At the level of the leaf or the plant, the metabolic process of primary interest is photosynthetic carbon dioxide fixation. Many of the pathways leading to the production of sugar and other carbon-containing compounds are common to both photosynthesis and respiration. Thomas[669] demonstrated that hydrogen fluoride fumigation reduced the rate of photosynthesis. The initial reduction was greater than the amount of leaf area damaged, but after a lag period the reduction of photosynthesis was equivalent to the reduction of leaf surface. The extent of the lag depended on the concentration of fluoride, duration of exposure, and species of plant. Thompson *et al.*[675] found no effect of low levels of fluoride on photosynthesis of bearing citrus trees, and Hill *et al.*[277] reported no effect of hydrogen fluoride on net photosynthetic rates of tomato during a 3- to 4-week fumigation period at relatively high concentrations.

It is generally assumed that foliar fluoride absorbed from the atmosphere remains in the inorganic form, except in a few exotic species that synthesize fluoro-organic compounds. However, Lovelace *et al.*,[378] Yu and Miller,[736] and Cheng *et al.*[138] have reported that mixed pasture forage, crested wheat grass, and soybeans exposed to atmospheric fluoride or hydrogen fluoride synthesize highly toxic monofluoroacetic and monofluorocitric acids. Although Lovelace *et al.*[378] reported a concentration of over 1 mg of fluoro-organic acid per gram of dry weight, a more recent publication[736] suggests that only a trace amount occurs–which would be in keeping with the earlier conclusion of Wade *et al.*[697] that, although microgram quantities of organic fluoride compounds were found, the concentrations were far too low to isolate and identify or to cause concern over toxic materials.

McCune and co-workers[431,432] have summarized present knowledge of the metabolic effects of fluoride on plants. Part of the discussion by McCune and Weinstein[431] follows (reference numbers conform to the citations of the present document, rather than of the original):

> The biochemical effects induced by fluoride that are not associated with chlorosis or necrosis are diverse and suggest that many different areas of metabolism can be affected. However, many of these effects have certain features in common. There is a threshold value of HF for each crop, depending upon duration of exposure and HF concentration, below which fluoride does not affect the metabolism of the plant,[277,669] Above this threshold the metabolic response has both a magnitude and a duration, and the magnitude of the response may decrease or become reversed with the cessation of fumigation[433,669,709] or with time during long-term fumigations.[7,40] The translocation[669] or inactivation of fluoride and the nonrate-limiting character of a fluoride sensitive step or pathway could explain the existence of a threshold. The apparent recovery could also be explained by the removal of fluoride from a sensitive site. The changes in enzyme levels and glucose metabolism that occur with age or fluoride treatment suggest that recovery may also

involve the development of fluoride insensitive pathways or inductive phenomena in glycolysis.

Fluorides may act as metabolic inhibitors, and the translation of the presence of the pollutant in the cell into an altered metabolic pattern may consist of the following sequence of events. The pollutant reacts with a metabolically active site and renders it inactive. Presumably this site is an enzyme although coenzymes or activators may be targets of the pollutant. The inhibition of the enzyme results in an inhibition of the reaction mediated by that enzyme. The inhibition of one step in a pathway leads to decreased activities in the subsequent steps and in the metabolic systems that this pathway supports.

There are certain qualifications of this skein of events which must be made, however. The degree of inhibition depends upon the concentration of inhibitor at the active site, and this in turn depends upon the amount and distribution of the inhibitor within the cell. Usually in a series of steps that constitute a metabolic pathway, one step is rate-limiting and acts as a pacemaker. If the pollutant-sensitive step is not the pacemaker, its inhibition will not affect the pathway until the degree of inhibition makes it the rate-limiting step. The inhibition of one step or a sequence of steps may not result in an appreciable decline in metabolic activity if alternate pathways exist or are activated as a result of the inhibition. It is evident that factors residing in the plant or present in its environment will affect the sensitivity of the plant to an air pollutant if they alter the sensitivity or availability of the active site to the pollutant, the relative activity of that site to the activity of the pathway, or the presence or functioning of alternate pathways.

As yet, we cannot translate these effects on metabolic processes into gross effects on intact organisms. However, some effects of fluoride on total growth, growth habit, and occurrence of symptoms are known.

Visible Symptoms

Vegetation exposed to an excessive amount of gaseous fluoride will develop symptoms of injury. The symptoms, although characteristic, are not necessarily specific.[93] Therefore, the review of fluoride-induced symptoms is followed by a review of mimicking symptoms induced by a variety of other agents.

Most of the general reviews provide some description of the typical symptoms associated with atmospheric fluoride.[93,239,278,670,681] Perhaps the best available summary of symptomatology is that by Treshow and Pack.[681] Their description is quoted here in part (reference numbers conform to the citations of the present document, rather than of the original):

> *Leaves* Fluoride air pollutants enter the plant primarily through the leaves. Most of it apparently enters through the stomata, passes into the intercellular spaces and is absorbed by the mesophyll.[673] From the mesophyll the fluoride

may move to other cells by simple diffusion or through the vascular tissue. It moves with the transpiration stream toward the leaf tips and margins where it accumulates in concentrations at least several times higher than the average concentration in the leaf as a whole.[742]

The characteristic symptom of fluoride injury on many broad-leaved plant species is necrosis, which occurs predominantly at the leaf tips and margins where the fluoride has accumulated. A dull, gray-green, water-soaked discoloration of tissues along the leaf tip and margin is the first sign of injury to appear on such sensitive species as apricot and European grape. . . . This symptom may appear within 24 hrs of a fumigation with fluoride concentrations of several ppb or, more typically, after several days or weeks, if concentrations are lower. These "water soaked" tissues generally turn light to dark brown within 48 hrs of fumigation during hot weather. Lower temperatures tend to delay symptom expression as much as several days. In the field, symptom expression may be delayed several weeks after the critical threshold of fluoride accumulation has been reached if temperatures remain cool and no further stress is imposed on the plant. If the atmospheric concentrations are extremely high fluoride may be absorbed by leaves at a much faster rate than it is translocated. Under these conditions, lethal concentrations apparently build up at various locations in the leaf tissue producing scattered intercostal lesions. . . . The scattered distribution of the lesions, and the more frequent presence of chlorotic areas, are the distinguishing characteristics of this type of exposure.

Apricot leaves exposed to low fluoride concentrations first develop semicircular lesions 1-3 cm in diameter along the leaf margin. A narrow, sharply-defined, reddish-brown band less than 1 mm wide typically separates the necrotic tissue from the adjacent healthy tissue. Microscopic examination has shown that this band often consists of an abscission layer formed by cell division. The necrotic tissue soon breaks loose along this band and drops off, so that unless subsequent markings develop, the only remaining sign of the injury may be the irregularity of the leaf margins. When fluoride concentrations are higher, or the period of exposure is prolonged, larger areas of tissue are affected and injury continues to develop. Generally fluctuations in atmospheric fluoride concentrations, or in environmental conditions that influence development of injury, cause the necrosis to appear in successive waves. A narrow, dark, reddish-brown band produced by deposition of resins and tannins in the peripheral cells delimits and characterizes the necrosis formed by successive fumigations and gives the necrotic lesions a wavy, zonate appearance. The necrotic tissue may ultimately form a continuous band around the leaf affecting up to 100% of the leaf or may be limited to trace amounts at the tip or along one edge.

Although the necrotic tissue readily separates and may drop out, the apricot leaf itself rarely abscises, even when the fluoride levels in the leaves exceed several hundred ppm and the leaf is severely marked. However, defoliation has been reported on certain other stone fruits and citrus.[101,175]

The necrotic lesions described for apricot are typical of the injury produced on sensitive, broad-leaved plants. The wavy color pattern of the necrosis is the most definitive characteristic, but the amount of color contrast varies among species as does the color of the necrotic tissue. Some species, *e.g.* boxelder, show little tendency for the necrotic tissue to break away from the normal tissue at its juncture, but it may become shattered and broken off in irregular pieces by wind.

The most sensitive leaves on fruit trees such as apricot and prune are those on sucker shoots with their softer, rank growth. Axillary and terminal buds are highly resistant to fluoride and trees have been observed to retain their capacities to produce new shoots for 10 successive years. . . .

Fluoride injury symptoms on needles of pine and other conifers consist of necrosis which begins at the tip of the current year's needles and progresses toward the base. . . .Injured tissue first becomes chlorotic and then turns buff to reddish-brown, depending on the species. Occasionally [rarely], bands of tissue part of the way down the needle become necrotic. Needles are most sensitive when they are elongating and emerging from the "fascicle" in the spring, and become progressively more resistant as the season progresses. Needles formed during preceding years are highly resistant to fluoride and are rarely, if ever, injured by current fumigations.

Solberg *et al.*[618] conducted detailed histological studies of the effects of fluoride on pine needles and observed that the tissue collapsed only a few cells in advance of the necrotic areas. Epidermis, hypodermis and xylem tissues were most resistant and parenchyma most sensitive. Phloem cells were the first to be injured. Phloem and xylem parenchyma, together with transfusion parenchyma cells, enlarged greatly, becoming extended and distorted. The protoplast then became granulated, vacuolated and finally collasped. These characteristics were useful in distinguishing fluoride-induced needle necrosis from that caused by sulfur dioxide and other agents.

Fluoride may cause a chlorotic symptom expression as on leaves of citrus . . . poplar . . . and sweet cherry. The chlorosis, or yellowing, extends inward from the leaf margins between the larger veins. In its early development, the chlorosis is light yellow-green in color and shades gradually into the normal dark green tissue along the midrib and larger veins. With continued exposure to fluoride, the chlorotic tissue may become completely yellow, and the demarcation between chlorotic and green tissue becomes sharply defined. Severe defoliation has also been reported to accompany this stage of leaf injury.[108] Some of the more severely injured tissue at the leaf margins may eventually become necrotic. Chlorosis has been observed to be most common on many other plants when fumigated with hydrogen fluoride in greenhouses and growth chambers. It does not appear to be common in the field, but occasionally is associated with leaf necrosis in plant species on which necrotic symptoms normally predominate.

Symptoms of fluoride injury on monocotyledonous plants are essentially similar to those on broad-leaved species. On gladiolus, necrosis develops primarily at the leaf tips, and reddish-brown waves denoting successive fumigations are striking and definitive. . . . Similar "bands" are also characteristic of fluoride toxicity on False Solomons-seal (*Smilacina*, sp.) and other sensitive species. . . . The zonation is absent only when the necrosis is caused by a single fumigation. The necrosis frequently extends further down one side of the leaf than the other. Sensitive varieties [of] iris (*Iris*, sp.), narcissus, tulip, and related species develop similar injury symptoms. Necrosis on cereal crops such as barley is light in color and sometimes almost white. These species are more resistant to fluoride, and necrosis observed in the field is more frequently due to hot, dry winds or other environmental stress.

Fluoride injury symptoms on corn consist of a chlorotic mottle, or "flecking," concentrated toward the margins and tips of the leaves. . . .[282] Small, irregularly-shaped, chlorotic spots develop between the veins gradually merging into continuous chlorotic bands. When injury is more severe, most of the leaf may become chlorotic. The yellowish to straw-colored chlorotic areas tend to be confined between the larger veins and concentrated along the outer edge of the interveinal zone. Tissues along the outer side of the large veins normally remain green, but the chlorosis may bridge these veins as it intensifies. When the injury is severe, some of the chlorotic tissue may become necrotic, particularly along the margins and at the tips of the leaves.

Fluoride injury symptoms on sorghum are similar to those on corn, but in fumigation experiments some varieties have shown a tendency to develop dark, brownish-red spots at the leaf tips and margins. Spots, 0.5 cm or more in diameter, may become necrotic in the center or smaller spots may fuse together and become necrotic but the tissue in scattered small spots generally remains turgid.

Flowers and Fruit Flower petals are rarely injured in the field. The only reported observations of injury are cited by Thomas[669] who lists petunia (*Petunia*, sp.) petals as intermediate in sensitivity, and Spierings, [622] who reported that cyclamen (*Cyclamen*, sp.) flowers were more susceptible to hydrogen fluoride than the leaves. Injury consisted of marginal necrosis of the petals and sepals. Gladiolus flowers are resistant to fluoride despite the extreme sensitivity of the leaves and bracts. Sepals and petals remain unmarked even when 50% or more of the leaf area is necrotic and the bracts subtending the flowers are equally damaged. [Note: Guderian *et al.*[239] present a color plate of flower injury on gladiolus. This reference was not available to Treshow and Pack.] Apple, apricot, cherry, and peach petals are also tolerant.

Some fruits may be indirectly more sensitive to fluoride than the leaves. The best known example of fluoride injury to fruit is the soft suture disease of peach. . . . This syndrome may be better termed "suture red spot" since this denotes a specific type of soft suture associated with fluoride,[68,545] and is characterized by premature reddening and ripening of a local, rather sharply-delimited area along one or both sides of the suture line toward the lower (stylar end) third of the fruit. The red area contrasts sharply against the pale yellow or yellow-green background color. In some instances, premature ripening and swelling cause a local enlargement of the affected area. More often the tissues along the suture ripen without undergoing excessive enlargement, and the affected portion is typically over-ripe, soft, and slightly sunken by the time the rest of the fruit is ripe. Premature reddening may appear 2 weeks to 1 month before harvest. The early, sharply-defined coloring, even on fruits in shaded parts of the tree, is especially striking before the normal ripening coloration begins. Another symptom is the separation of the flesh along the suture. This splitting beneath the skin may occur even when symptoms are extremely mild. Reddening of the affected flesh in a local region along the suture is another good diagnostic aid with yellow-fleshed varieties such as Hale and Elberta.

Fluoride-induced suture red spot may be influenced by calcium and boron nutrition, and it has been a problem in certain areas where atmospheric fluoride concentrations have been very low. Even where fluoride concentrations were too low to cause necrosis of peach leaves, up to 90% of the fruit was rendered unsaleable. Suture red spot is not correlated with high fluoride concentrations in the fruit since the critical period of sensitivity appears to be early in fruit development when the pit is hardening and calcium demands are greatest. At maturity, peaches, like most fruit, never contain more than a few ppm of fluoride.

Less striking but basically similar symptoms attributed to fluoride have been described and illustrated by Bolay and Bovay[83] to consist of a local necrosis of the stylar (*i.e.*, basal) end of apricot, cherry and pear fruits. A round or irregular sunken, dark brown to black lesion developed at the stylar end of the fruit as it ripened. This symptom has been noted on apricot fruits in two Utah orchards under conditions of moisture stress and poor nutrition. Although the fluoride levels of leaves in these orchards exceeded 200 ppm the relation of fluoride to symptom expression was not known. No symptoms appeared on pear fruits in the same orchards.

A similar condition known as "snub nose" or "shrivel tip," in which the stylar end, or tip, of the cherry fruit ripens and shrivels a few days prematurely, has been observed on the Orb variety. The disorder may be caused by high fluoride concentrations alone, but is aggravated by water stress; almost indistinguishable symptoms can be caused by water stress alone. A "black tip" condition of sour cherries and peaches is also attributed largely, if not wholly, to water stress.

Mimicking Symptoms

Several diseases and environmental stresses may produce visible changes in leaves that are very similar to, if not identical with, those produced by excess fluoride in the tissue. Treshow and Pack[681] summarize the most common sources of confusion in identification of fluoride injury as follows:

> Diagnosis of fluoride injury occasionally may be complicated by the presence of symptoms which look similar to those produced by fluoride but whose cause is completely unrelated to fluoride pollution. Accurate diagnosis depends on familiarity with the normal appearance of plants and knowledge of the relative fluoride sensitivity of a wide range of species. The observer must be familiar with symptoms caused by other agents as well as the symptoms of fluoride injury. The more he knows about the local disease and injury problems caused by fungi, viruses, bacteria, insects, mites, improper nutrition or culture, temperature and moisture stress, and other "normal" environmental stresses, the more accurate and valid will be his diagnosis and evaluation.

Fluoride analysis is often a useful aid to diagnosis. For such data to have greatest value though, the observer must have some knowledge of what constitutes "normal" background levels. In the absence of an atmospheric fluoride source, plants commonly accumulate 5–10 ppm fluoride in their leaves during the course of a growing season, and normal levels of 10–20 ppm are not unusual. A few species, however, included [*sic*] tea (*Thea*, sp.), camellia and gifblaar [*Dichapetalum cymosum*], have a unique capacity of accumulating vast quantities of fluoride, and foliar concentrations may reach several hundred ppm in the absence of atmospheric pollution.[673]

If the fluoride levels in an area being studied are "normal" fluoride can be ruled out as the injurious agent. Unfortunately, the reverse doesn't hold. The presence of higher fluoride concentrations in plant leaves does not prove that the fluoride is injurious, but such data may give some indication if the relative sensitivity of the plant is known. Similarly, partly because of the lack of data denoting the direction or source of exposure, and partly because of the multitude of environmental factors influencing sensitivity, atmospheric fluoride analysis provides only a general guide as to the possible presence or threat of fluoride injury.

Diagnosis of suspected fluoride injury to vegetation should begin by the location and careful examination of sensitive, indicator species in the area. The more closely the degree of injury is related to the fluoride sensitivity of various species, the more confidence can be placed in the diagnosis. If leaf markings are confined to resistant or tolerant species, some other causal agent should be sought.

Several environmental stresses cause injury symptoms strikingly similar to those characterizing fluoride injury. Fluoride injury on many plants is more closely mimicked by marginal leaf burning caused by moisture stress. . . . Such symptoms may be misleading to the untrained observer when they appear on such fluoride-sensitive, indicator plants as apricot. . . . Moisture stress causes marginal necrosis of many broad-leaved plant species, especially when cool spring weather has induced soft, rank growth which is especially predisposed to injury. Drought-induced necrosis can often be distinguished by a rather diffuse zone of chlorotic tissue separating necrotic and healthy tissues. Low-temperature injury, consisting of necrotic leaf margins may also resemble fluoride injury. . . .

Necrosis of conifer needles . . . is caused by so many stresses, and is so non-specific, that the total syndrome must be relied upon for diagnosis. The relative fluoride sensitivity of the affected and associated species is also helpful. Necrosis caused by other pollutants, especially chloride and sulfur dioxide, may also resemble fluoride injury not only on conifers, but on such fluoride-sensitive broad-leaved species as European grape. . . . Again, the relative sensitivity of different-aged tissues and different species must be considered.

Symptoms of virus and genetic disorders such as prune leaf spot and grape leaf roll may at times resemble fluoride injury . . .; therefore several different plant species should be examined for symptoms whenever possible.

Pesticides, especially oil sprays, organic phosphates, and other acutely toxic chemicals can cause necrosis similar to that caused by fluoride. . . .

Some nutrient deficiency symptoms bear a close resemblance to fluoride-induced chlorosis. The marginal and interveinal chlorosis that characterizes manganese deficiency on plants such as peach, cherry, and citrus . . . is so similar to mild

> fluoride toxicity symptoms that one must rely heavily on the appearance of symptoms on associated species. Zinc deficiency symptoms on leaves may also be confused with fluoride injury unless the total syndrome is evaluated and fruits can be inspected. The malformed fruit, leaf distortions, and rosetting associated with zinc deficiency are not associated with fluoride injury.
>
> Symptoms of fluoride toxicity on corn are often similar to those produced by zinc or potassium deficiency, mite injury, genetic variation, or normal senescence. Generally the genetic mottle is brighter yellow in color. Similarity of fluoride injury to normal senescence renders diagnosis late in the growing season tenuous.
>
> Several pathogenic agents can cause local, premature reddening and ripening of the suture of peach fruit, and the general name soft suture has been given to this expression regardless of cause. In all cases the affected area colors and ripens 2 or 3 days early.
>
> A physiological type of soft suture characterized mostly by stress-related, early ripening of one side of the fruit was described and named soft suture by M.H. Dorsey in Illinois in 1944.[180] In constant contrast to the fluoride-induced soft suture, which is usually more prevalent on trees with a light crop, the condition was associated with overloaded, weak trees.
>
> Growth-regulating chemicals such as the phenoxyacetic acids can also cause a soft suture expression when applied in excessive amounts to improve fruit size or to control weeds in orchards while the fruits are developing. . . . In contrast to fluoride-induced injury, symptoms are most pronounced near the stem end of the fruit and the swelling is more pronounced.

Growth, Yield, and Quality

Scurfield[577] included some observations on fluoride in his discussion of the effects of air pollution on tree growth. The report of the 1961 Congress on Smoke Damage to Forestry, held at Tharandt, Germany,[689] gives further general reviews with an emphasis on sulfur dioxide and dusts. Weisser[716] discussed some of the general problems of tree survival in polluted areas with only limited specific reference to fluoride.

Lynch[383] identified a reduced growth rate in the "pine blight" area near Spokane, Washington, as a direct effect of fluoride.[581] Adams *et al.*[13] reviewed the extent of the growth reduction on pine in the Spokane area and associated it with development of necrosis in the needles. There was no evidence of a significant reduction in growth without necrosis. Wentzel[718,719] reported on growth reduction in trees around fluoride sources (brick plants) and on recovery in growth rates associated with reduction in emissions; growth reduction was accompanied by foliar symptoms.

Treshow *et al.*[679] presented evidence of a significant growth reduction in Douglas fir as a result of fluoride, in the absence of necrosis of the needles, but they also reported an increase in needle size associated with increased fluoride content. Growth reduction of some species can occur as a result of fluoride without characteristic foliar symptoms of chlorosis and tip burn. Use of hydroponic culture with added fluoride suggested that leaf size on some citrus may be reduced at leaf concentrations of 50–60 ppm.[100,243] Brewer and co-workers,[101,102,105,106,108] using fumigations with hydrogen fluoride and sprays of various alkali fluorides, concluded that chlorosis, reduced leaf size, reduced growth (judged by several criteria), and reduced fruit yield can result from exposure to and uptake of fluoride. The studies of Leonard and Graves[361] in Florida agree with the observations from California.

Because citrus holds its leaves for relatively long periods and produces several flushes of new leaf development and growth each year, a threshold of leaf fluoride concentration that can be related to an effect is difficult to define. The available data suggest that concentrations of 200 ppm in mature leaves are associated with significant reductions in yield and total growth. Comparable effects have been associated with 100 ppm in fully expanded new flush leaves. A threshold would be considerably lower. Some of the hydroponic culture work,[100,243] as well as limited observations in field and fumigation trials,[101,102,108] has suggested a possible threshold of fluoride for growth effects in the range of 50–100 ppm.

For a discussion of the problems involved in relating fluoride damage to accumulation in foliage, see the reports of Bolay and Bovay[83] and Schneider.[573,574]

Hill *et al.*[277] concluded that hydrogen fluoride had no effect on growth of tomato, even at fluoride concentrations that produced slight necrosis. Applegate and Adams[41] and Pack[501] concluded that a significant decrease in growth and fruiting of bush beans was unlikely in the field, although Pack[502] and Stocks[631] produced significant growth reduction in beans at high fluoride concentrations.

An abstracted paper by Kazantseva[329] reports significant reduction in grass yields near an aluminum smelter in Russia. If the reported air analyses for fluoride are correct (up to 0.612 mg/m^3), this is not too surprising. The abstract contains no data on the fluoride content of the grass.

A report on the uptake of fluoride from soils treated with sodium fluorosilicate[257] suggests that the yield of alfalfa was reduced when the content of fluoride in the tops exceeded 60 ppm. This example does not represent a common field problem. The reduced growth may be a reflection of very high fluoride content in the roots, rather than the moderate fluoride content in the tops. The data from Treshow and Harner[680] suggest stimulation of growth, or at worst no effect, in alfalfa at concentrations of up to 200 ppm.

The reports of growth stimulation by fluoride are not new. In 1905, Aso and Suzuki[48] reported that applications of 80 g of sodium fluoride per hectare increased the yield of rice; application of 800 g/hectare had no effect. More recently, Adams and Sulzbach[16] reported that low-level fumigation with hydrogen fluoride increased the internode length of beans. Treshow *et al.*[679] reported that the needles of Douglas fir exposed to fluoride were larger than unexposed controls. Both O. C. Taylor of California and D. F. Adams of Washington observed growth stimulation due to fluoride (quoted in Treshow and Harner[680]).

Treshow and Harner[680] exposed beans and alfalfa to hydrogen fluoride at fluoride concentrations ranging from 0.3 to 3.2 $\mu g/m^3$. Beans were exposed for 3 weeks, alfalfa for 4 weeks. A number of paired groups of plants were exposed to filtered air and to various higher concentrations. Plants were harvested, the fresh weight and dry weight were determined, and the tissue was analyzed for fluoride. The plant weights were arranged in groups according to fluoride content. Control bean plants with a fluoride content of less than 50 ppm had an average fresh weight of 9.0 g and a dry weight of 1.03 g. Bean plants with fluoride contents of 51–100 ppm and 101–150 ppm had fresh and dry weights not significantly different from those of the controls. However, beans with fluoride contents of 151–200 ppm had a fresh weight of 13.9 g and a dry weight of 1.88 g. The results were significant at the 5% level of probability (*t* test). The group with fluoride contents of 201–300 did not differ from the controls. The fresh and dry weights (5.4 and 0.63 g) of the group with fluoride contents in excess of 300 ppm were significantly less (5% level of probability) than those of the controls. Similar trends were noted with alfalfa, but the differences were not statistically significant. The results for alfalfa were more suggestive of reduced growth at higher fluoride concentrations in the tissue (above 200 ppm) than of stimulation at lower levels.

Fluoride can reduce flower quality and corm production of gladiolus.[103,281] This reduction reflects the degree of leaf injury. Similar reductions can be induced by clipping leaves.[149] There is no evidence of any growth reduction without tip burn on gladiolus.

Corn and sorghum may react differently.[280,282] In both crops, reduced yield can often be correlated with foliar symptoms. However, when fumigations occur at specific stages of tassel development, grain yield can be significantly reduced without chlorosis or other leaf symptoms. The atmospheric concentrations of fluoride that reduce the yield of corn or sorghum, with or without chlorosis or necrosis, are within the range of values associated with field problems.

Deleterious effects in fruits, however, do not necessarily occur only during the flowering period. The condition known as "suture red spot" or "soft suture" of peaches has been associated with atmospheric fluoride,[68,681] and fruits are believed to be most susceptible shortly before pit hardening. Direct effects of fluoride have also been reported in apricots, pears, and cherries and were characterized by the appearance of depressed lesions at the stylar end of the fruit.[83] Treshow and Pack[681] have also described a condition called "snub nose" or "shrivel tip" of sweet cherry, which has been ascribed to a high atmospheric concentration of fluoride.

Most of our information on the characteristics of exposure that relate fluoride to specific plant responses is derived from experimental fumigations in which plants were exposed to known concentrations of hydrogen fluoride for different periods. Unfortunately, the amount of information available is limited, for technical and other reasons, to (1) relatively short exposure periods, (2) the species and varieties of plants tested, and (3) an elementary knowledge of the influence of climatic and edaphic factors on plant growth and reproduction. It is difficult to compare results between laboratories because of differences in such factors as equipment, and methods of analysis. Furthermore, the credibility of these experiments in relation to industrial pollution is reduced by the experimental difficulties of reproducing the temporal distribution of fluoride and variations in climatic and edaphic conditions, although these difficulties are becoming surmountable. Thus, discussion of the effects of fluoride on plant growth and reproduction has been limited primarily to greenhouse- and field-grown plants exposed to gaseous hydrogen fluoride within the confines of a fumigation chamber.

Growth may be defined as an irreversible increase in mass or (as by some botanists) as a change in any dimensions, including length, surface area, or volume. A number of dimensional changes have been associated with atmospheric fluoride, including changes in radial growth (a decrease) of Douglas fir[679] and citrus;[101,105,108] dry weight of aboveground portions of roses and citrus;[105,107,108] weight of stalks of sorghum;[282] leaf size and area;[101,105,108,358] weight of aboveground parts of alfalfa, orchard grass, and romaine lettuce;[67] number of flowers or fruits of tomato, citrus, sorghum, and bean;[102,282,361,503] and weight of roots.[67] These published effects represent a broad range of atmospheric fluoride concentrations, durations of exposure, times of exposure in relation to plant development, and climatic and edaphic factors. For example, reports of effects on Douglas fir are based on field observations with no air analysis; effects on citrus, on both field and greenhouse experiments carried out over relatively long periods; effects on sorghum, on observations of short duration, mostly during heading; and effects on bean and tomato, on long-term exposures in controlled greenhouse conditions. It is not surprising, therefore, to find reports of observations that fluoride exposures have not induced significant effects on growth and development—e.g., in sorghum,[282] corn,[280] gladiolus,[281] spinach, endive, orchard grass, alfalfa, chard, romaine lettuce,[67] tomato,[277] bean,[503] and lemon.[675] Indeed, there is evidence that exposure to fluoride may stimulate growth, as has been reported for Douglas fir needles and shoots,[679] total linear growth of citrus and roses,[101,107,108] longer internodes in beans,[16,42] seed head production in sorghum,[282] and growth of alfalfa.[680] Although, in some limited circumstances, an actual growth enhancement may occur, it would be naive to accept all this information as evidence of the beneficial effects of fluoride on plants, because they can be more appropriately ascribed to a more twiggy growth in roses and citrus and taller but not necessarily sturdier bean plants. Fluoride was recently reported to be an essential trace element for the growth of corn,[73] but only in special circumstances.

Accumulation of Fluoride

Fluoride is present in various amounts in all soils and natural waters. There is no reason to expect clean, unpolluted air to be completely free of fluoride. All vegetation contains some fluoride. The primary source of natural fluoride in vegetation is the soil. Irrigation water could in some cases add a significant amount, and air that is apparently uncontaminated might contribute a minute amount.

Because our interest has been in the toxic amounts of fluoride (toxic either to the plant or to the animal that consumes the plant), it is not surprising that more data are available on increased fluoride contents of vegetation than on the normal content. Garber[218,219,222] presented the most complete list of normal fluoride contents for a variety of plants grown in several soils.

The natural concentration of fluoride in the foliage of most plants is in the range of 2–20 ppm (micrograms of fluoride per gram of dry weight). The value varies with age of leaf, soil, species, and even variety.[218,219,222] Some plants (the family Theaceae–teas, camellias, etc.) seem to accumulate fluoride from soil and water. Normal values for these species range from 50 to several hundred parts per million. The fluoride concentrations of 107 samples of alfalfa from different areas of the United States assumed to be free of industrial pollution ranged from 0.8 to 36.5 ppm, with a mean of 3.6 ppm.[641]

Although the soil is undoubtedly the principal source of the normal fluoride content of plants, there is ample evidence that there is no relation between the concentration in the soil and that in the plant.[6,220,222,256,257,317,388,528] Data indicate that the more acid soils generally result in plants of slightly higher fluoride content. The data are more specific in indicating that liming of acid soils usually reduces the fluoride content of the crop.

Several groups of workers have studied the effect of fluoride additions to soils on the fluoride content of the crops grown.[220,222,256,257,388,390–395] Their data show that the relatively insoluble fluoride compounds–e.g., calcium fluoride, rock phosphate, cryolite, slags–have little or no effect on fluoride uptake by vegetation when added to the soil, even at high rates of application. The soluble fluoride salts could increase the amount of fluoride taken up by plants. The reponse to soluble fluoride salts depends on the soil, as well as on the amount of added fluoride. The data of Hansen *et al.*,[257] who added sodium fluoride to four different Utah soils, are typical. With no added fluoride, alfalfa grown in the four soils contained about 15–18 ppm. With three of the soils, additions of fluoride up to 800 ppm resulted in only slight increases of fluoride in the alfalfa. Addition of 1,600 ppm markedly increased the fluoride content of the alfalfa and also significantly reduced the yield. In the soil of noncalcareous derivation–Orem loamy sand–the fluoride content of the alfalfa was almost doubled by the addition of 200 ppm to the soil. The yield was significantly reduced in this soil at 800 ppm. The conclusion of all these workers is that the amounts of fluoride added to soil (i.e., those incidental to general fertilizer practice) have little or no effect on the fluoride content of the plant. MacIntire and his co-workers[388,390,391,393] suggest that there is little or no possibility that industrial air pollution adds enough fluoride to soils for soil-derived fluoride in the crops to be significantly increased. Most of the workers who have critically reviewed the literature agree with this general conclusion.[6,93,218,669,671]

A few European workers suggest that soil contamination does occur, but at present there are no data to support their contention.[81,204,247]

There is little detailed information on the amount of fluoride in plant tissue derived from irrigation water. But there is ample evidence from hydroponic culture work of increased levels of fluoride in crops reflecting increased concentration of fluoride in the solution.[16,165,364,503] Several investigators who have used sprays containing soluble fluoride salts indicate that they can be absorbed directly through leaves.[68,102,105,106] Rand and Schmidt[532] have reported an increased fluoride content of forage grown with irrigation water containing 6.2 ppm of fluoride. This suggests that irrigation waters high in fluoride could add a small amount of fluoride in plants. However, the problem should be marginal and of very local concern.

Some workers have been rather pessimistic about our ability to relate atmospheric fluoride concentrations to fluoride uptake by a growing plant over a reasonably long period.[425,681] McCune,[424,425] in a survey of the available literature, suggested criteria relating atmospheric fluoride concentrations to growth response and development of foliar symptoms. In his discussion of alfalfa, he emphasized the importance of relating plant content to air concentration, but suggested that it had not yet been possible to establish a relation. Treshow and Pack[681] reached a similar conclusion.

The fluoride content of forage plants growing in a fluoride-contaminated environment is important. Criteria based on the fluoride content of animal diets have been suggested.[519,585,592,638] If the forage levels exceed these dietary criteria by a factor of 2 in an existing area of contamination, it is obvious that the air concentration of fluoride should be reduced by a similar factor or more. The actual air concentration need not be known. The engineer concerned with emission control and the official concerned with air-resource management will take justifiable exception to such a simplification of the problem. Some estimate of the relation between average air concentration and uptake of fluoride by forages is needed, even though the estimate may have a high degree of uncertainty. For a discussion of this problem, see Hendrickson.[268]

If a plant were a simple passive accumulator of fluoride from the air, one might expect the increase in fluoride concentration to be proportional to the atmospheric concentration:

$$dF = KC\,\mathrm{d}T,$$

where F is the concentration of fluoride in the plant, C is the concentration of fluoride in the air, and T is the period of exposure. This assumption leads to the often-cited dose–rate relation:

$$\Delta F = KCT. \tag{1}$$

Because there is a baseline fluoride content of plants from nonatmospheric sources, ΔF represents the difference in fluoride content attributable to atmospheric fluoride.

Except for short periods, we should not expect plants to accumulate fluoride passively. Stomata open and close; leaf areas and surface-to-mass ratios change. All these changes obviously affect the coefficient K. Other changes related to growth and environmental factors are likely to affect the value of K. Such factors as wind speed could have an enormous effect on the apparent value of K, even if the leaf surface were a consistent, passive accumulator.

Despite the obvious limitations, the simple dose–rate relation (Eq. 1) may be useful. However, it is necessary to recognize and accept the fact that the apparent accumulation coefficient, K, is not a constant.

One should bear in mind that most experimentally derived coefficients are based on controlled fumigation studies and relate only to the effects of hydrogen fluoride. Thus, the effects of particulate fluoride in the atmosphere must also be considered when accumulation of fluoride by forage is important.

The differences in tolerance to fluoride in plant tissues, as indicated by a visible foliar response (necrosis or chlorosis), are very great.[317] Some varieties of gladiolus will develop necrosis when the tissue fluoride concentration exceeds 20 ppm, whereas cotton may show no injury at more than 4,000 ppm in the leaf tissue. Brandt and Heck[93] suggested that plants can be grouped into two classes: plants that develop leaf symptoms when the fluoride content in the tissue is 50–200 ppm and plants that show no injury above 500 ppm. Hill[275] suggested a range of 20–200 ppm for probable injury to susceptible plants. Treshow and Pack[681] reported that there is a highly susceptible group of plants that shows injury at a tissue concentration of less than 50 ppm.

The threshold for injury for all plants listed as susceptible in the various published lists is certainly less than 150 ppm. For most plants so listed, it may be less than 100 ppm.[82,86–88,221,239,669,681,742] Plants classified as intermediate or resistant can probably all tolerate fluoride accumulations in excess of 200 ppm without developing chlorosis or necrosis.

The data on growth reduction in relation to fluoride content are limited. Data for bean and alfalfa[680] suggest no effect on either crop up to 200 ppm, but bean growth is reduced when the content exceeds 300 ppm. Data on citrus[101,102,108] suggest a possible threshold for growth effects in the range of 50–100 ppm, and 100 ppm is suggested as a threshold for growth reduction in Douglas fir.[679]

FACTORS AFFECTING PLANT RESPONSE

As important as the effects of fluoride on a plant are the conditions that determine the plant's response to it. One may group these factors as characteristics of the environment, of the biologic receptor, and of the pollutant itself. Each factor can influence the occurrence, type, and degree of effect and can exert its action before, during, or after exposure (with the exception of factors attributed to the pollutant itself).

Environmental Factors

Although the atmospheric concentration of fluoride, the duration and frequency of exposure, the age or stage of development of the plant, and the species and variety of the plant are the major factors associated with plant susceptibility, most climatic and edaphic factors also exert important effects on plant response.

Plants with a water deficit have been shown to be more resistant to fluoride than those receiving adequate water.[165,742] Light also plays a role in modifying the response of plants to fluoride. Plants fumigated in darkness are less injured than those fumigated in light.[165] However, in susceptible species, injury was shown to occur in both light and darkness at about the same foliar fluoride content, whereas injury in resistant species was observed in the dark when the foliar fluoride accumulation was only about half of that in the light.[8] Some investigators have reported that fluoride accumulation in the tissues of plants was greater in light than in darkness and concluded that uptake of fluoride into foliar tissues was lower in the dark because stomata were closed. They also demonstrated that the accumulation of fluoride increased in roots and decreased in tops with increasing root temperature.[66] These results suggest a downward translocation of fluoride from tops to roots at higher soil temperatures, which is contrary to previously published work.[65,358,362] MacLean and Schneider[398] have recently reported that gladiolus plants exposed to hydrogen fluoride at a constant vapor-pressure deficit exhibited greater tip burn with increasing temperature. Foliar accumulation of fluoride decreased in gladiolus but increased in sunflower leaves. Suboptimal nutrient supplies of calcium, magnesium, potassium, phosphorus, and nitrogen have been associated with increased severity of fluoride-induced markings.[16,98,397,430,503] It has also been shown that the amount of foliar injury on alfalfa foliage increased with the rate of air exchange in fumigation chambers.[279]

Pollutants themselves must also be regarded as environmental factors, because in ambient conditions they occur in various combinations. Little is known of the modifying influence of other pollutants in the presence of fluoride. Hydrogen fluoride or fluorosilicic acid in combination with hydrocarbons was found to produce less leaf marking on sorghum and gladiolus than hydrogen fluoride alone.[281,282] No synergistic or antagonistic effect between sulfur dioxide and hydrogen fluoride was found for several species.[281]

The Pollutant

The form of atmospheric fluoride influences its phytotoxicity. Gaseous forms may be the most important with respect to effects on vegetation, and the principal compound used in experimental fumigations is hydrogen fluoride. Fluorine, silicon tetrafluoride, and fluorosilicic acid have been used infrequently. The comparative effects of fluorosilicic acid and hydrogen fluoride were reported for gladiolus and sorghum by Hitchcock *et al.*,[281,282] who noted that fluorosilicic acid was more active than hydrogen fluoride in sorghum but equally active in gladiolus. Particulate fluoride compounds are usually much less toxic than the gaseous forms, and their toxicity is probably related to solubility[507] and to the size of hydrated ionic species.[106] The effect of particle size is not known, but cryolite particles 0.5 μm or more in diameter have not damaged plants.[429,507] The exact nature of the relation of degree of effect to the concentration of the fluoride in the atmosphere or duration of exposure is not known. The simplest assumption is that the degree of injury has the same relation to concentration and time as does fluoride accumulation.

In general, for the threshold of some effect, a rectangular hyperbola—$K = (C - C_t)(T - T_m)$—may describe the relationship between concentration and exposure, where one asymptote (C_t) would be called the "threshold concentration," and the other asymptote (T_m), the "minimal time" required for any concentration to produce an effect. Both these asymptotic values depend on many factors. For example, T_m would reflect the rate of diffusion from the atmosphere to the leaf surface and the penetration from the leaf surface to the interior cells. It would be a boundary condition determined mainly by physicochemical considerations. From a practical standpoint, this value would be hard to describe experimentally, because the concentration of fluoride required would be extremely high, and the time short. If we let T_m be equal to zero, the equation becomes that proposed by O'Gara[670] for sulfur dioxide injury. However, C_t, or the threshold concentration, is of great practical significance, because its determination would allow one to specify air-quality objectives in terms of concentrations that will produce no effect. This threshold concentration however, will depend on all the biologic and environmental factors.

Up to this point, we have assumed that we are dealing with continuous exposures to constant levels of fluoride. Unfortunately, the application of results from experimental fumigations to field problems is limited, inasmuch as constant concentrations over relatively long periods are not encountered in the field, and the effects of recurrent fumigation have not been studied, except for the work of Adams and Emerson[7] and MacLean *et al.*,[399] who found that the frequency or sequence of exposures was as important as concentration and time, although the former was concerned with metabolite levels in pine needles and the latter with fluoride accumulation in forage.

Biologic Factors

The stage of development or age of the plant is an important biologic factor, but when workers in the field of air-pollution effects on plants discuss stage of development, they may have two things in mind: They may be referring to a period in the life cycle of the plant, such as flowering, which is truly a stage in development if one considers annual or biennial plants, but not perennial plants, which repeat the cycle each year; and they may be referring to stages in the life cycle of an individual leaf, which is indirectly related to the life cycle of the parent plant.

Furthermore, in discussing the effects of fluoride on stage of development, researchers may be referring to two types of observations: In experimental fumigations, a plant or leaf may be exposed to the same dose of fluoride at different stages of its development; and in the field, a given amount of injury may be observed on foliage of different ages on the same plant or on plants examined at different times of the year.

Thus, if one is interested in yield information, it may appear that the best evidence on fluoride effects comes from controlled fumigations of sorghum and gladiolus. It was demonstrated that exposure of sorghum to fluoride during tassel-shooting and anthesis reduced the yield of seeds and aboveground portions of the plant. Exposures before and after this relatively short period in development had no effects on yield.[282] In part, the observed effect on yield may have been due to an effect of fluoride on stalk elongation or to an effect on leaf mass. Indirect effects on yield by direct effects on leaf mass have been demonstrated in gladiolus corms.[281] Let us assume that a given exposure will produce a known amount of necrosis of gladiolus leaves that will reduce corm yield in the mature plant. Plants exposed to fluoride at an earlier stage of development may exhibit the same percentage of necrosis after exposure as that required to reduce corm yield, but subsequent foliage will be unmarked and the necrotic percentage of total leaf mass would be proportionately less by the time of corm production. Exposure at a later stage of development or continued exposures during growth might result in destruction of leaf mass during the period of corm production sufficient to reduce yield.

For the most part, effects on susceptibility refer not to yield but to the presence or degree of leaf injury. It is generally assumed that immature leaf tissues are most susceptible to fluoride, as in the case of ponderosa pine, in which elongating needles are highly susceptible and mature needles are quite resistant.[15] This clear distinction, unfortunately, is blurred by the latency of some fluoride effects and their expression, and it is not unusual to observe the appearance of necrosis on fully elongated pine needles that have accumulated enough fluoride to be injured when environmental stresses occur later in the season.

For reasons not yet understood, plants exhibit an extremely broad range of tolerances to fluoride, as well as to other air pollutants. Great differences in susceptibility are found, not only between different species, but also between varieties of the same species. The resistance of camellia, cotton, and celery to fluoride may be contrasted with the extreme susceptibility of gladiolus, Chinese apricot, Italian prune, and developing needles of ponderosa pine. This differential response may also be found among varieties of, for example, gladiolus,[270,281] sweet corn,[280] and sorghum,[282] although the degree of difference is not as great. In gladiolus, dark-flowered varieties (purple and red) have been reported to be more resistant to fluoride than the lighter-flowered varieties (white, yellow, orange).[270] The white-flowered gladiolus variety "Snow Princess," for example, has been reported to be five times more susceptible to fluoride than the lavender-flowered variety "Elizabeth the Queen".[281] A similar difference has been observed among some conifers, such as ponderosa pine; the natural population appears to be homogeneous but is actually composed of a number of individuals having somewhat different genetic complements and showing great differences in response to fluoride.

Most frequently, specific and varietal differences in susceptibility have been tabulated on the basis of amount of injury produced by a given dosage of fluoride,[65,165,239,742] dose of fluoride required to reach the threshold for foliar markings,[8] and concentration of fluoride present in injured leaves.[82] These different means of estimating susceptibility account, at least in part, for the anomalous position of several species in these tabulations, although the lists are similar in most respects. Differences in varietal or specific susceptibilities may also be determined by the response actually observed—i.e., ratings according to leaf injury, linear growth of shoots, weight of tops, or size of fruit.[101,102,105,107,108]

SUSCEPTIBILITY AND RESISTANCE TO FLUORIDE

Perhaps the effects of genetic and other biologic factors may also be demonstrated when the nature of the susceptibility and resistance of plants to fluoride is considered.

There are many general lists of species arranged in order of susceptibility to fluoride.[82,86–88,221,239,363,669–671,681,712,740,742] Table 7-2 is a compilation for many North American plant species. Because the rankings are based on experimental and subjective field observations by different investigators, this list is intended as an approximate guide to the species shown, and it is not surprising that some are ranked differently in other lists.

The available information regarding susceptibility to fluoride using foliar lesions as the criterion suggests the following:

- Several of the conifers of importance to forestry, as well as ornamentals, are highly susceptible. Several more are in the intermediate class. One example of the confusion in classifying a

TABLE 7-2 Susceptibilities of Selected Plants to Fluoride[a]

Plant	Susceptible	Intermediate	Resistant
ACERACEAE			
Box Elder (*Acer negundo* L.)	—		
Maple, Hedge (*Acer campestre* L.)		—	
Maple, Norway (*Acer platanoides* L.)		—	
Maple, Silver (*Acer saccharinum* L.)		—	
AMARANTHACEAE			
Pigweed (*Amaranthus retroflexus* L.)		—	
AMARYLLIDACEAE			
Narcissus (*Narcissus* sp.)		—	
ANACARDIACEAE			
Sumac, Smooth (*Rhus glabra* L.)		—	
Sumac, Staghorn (*Rhus typhina* L.)		—	
BERBERIDACEAE			
Oregon grape (*Mahonia repens* Don.)	—		
BETULACEAE			
Birch, Cutleaf (*Betula pendula* var. *gracilis* Roth)			—
Birch, White (*Betula papyrifera* Marsh.)			—
CAPRIFOLIACEAE			
Elderberry (*Sambucus* sp.)			—
CARYOPHYLLIACEAE			
Chickweed [*Stellaria media* (L.) Cyrillo]	—		
Sweet William (*Dianthus barbatus* L.)		—	
CHENOPODIACEAE			
Goosefoot, Nettle-leaf (*Chenopodium* sp.)		—	
Lamb's Quarters (*Chenopodium album* L.)	—		

TABLE 7-2 Continued

Plant	Susceptible	Intermediate	Resistant
Spinach (*Spinacia oleracea* L.)			—
COMPOSITAE			
Aster (*Aster* sp.)		—	
Burdock (*Arctium* sp.)			—
Chrysanthemum (*Chrysanthemum* sp.)			—
Dahlia (*Dahlia* sp.)		—	
Goldenrod (*Solidago* sp.)		—	
Sunflower (*Helianthus* sp.)		—	—
CONVOLVULACEAE			
Potato, Sweet (*Ipomoea batatas* L.)	—	—	
CORNACEAE			
Dogwood (*Cornus* sp.)			—
CRUCIFERAE			
Cabbage (*Brassica oleracea* L.)			—
Cauliflower (*Brassica oleracea* L.)			—
CUCURBITACEAE			
Cucumber (*Cucumis sativus* L.)			—
Squash, Summer (*Cucurbita pepo* L.)			—
ELEAGNACEAE			
Russian olive (*Eleagnus angustifolia* L.)			—
ERICACEAE			
Azalea (*Rhododendron* sp.)	—	—	
Blueberry (*Vaccinium* sp.)	—		
Laurel, Mountain (*Kalmia latifolia* L.)			—
Rhododendron (*Rhododendron* sp.)		—	

TABLE 7-2 Continued

Plant	Susceptible	Intermediate	Resistant
FAGACEAE			
Oak (*Quercus* sp.)			—
GERANIACEAE			
Geranium (*Geranium* sp.)		—	
GRAMINEAE			
Barley (young plants) (*Hordeum vulgare L.*)	—		
Barley (mature plants) (*Hordeum vulgare* L.)		—	
Cane, Sugar (*Saccharum officinarum* L.)			—
Corn, Sweet (*Zea mays* L.)	—	—	
Crabgrass (*Digitaria sanguinalis* Scop.)	—		
Johnson Grass [*Sorghum halepense* (L.) Pers.]		—	
Oat (young plants) (*Avena sativa* L.)		—	
Oat (mature plants) (*Avena sativa* L.)		—	—
Rye (young plants) (*Secale cereale* L.)		—	
Rye (mature plants) (*Secale cereale* L.)		—	—
Sorghum (*Sorghum vulgare* Pers.)	—	—	
Wheat (young plants) (*Triticum* sp.)		—	
Wheat (mature plants) (*Triticum* sp.)		—	—
HAMAMELIDACEAE			
Sweet Gum (*Liquidambar styraciflua* L.)			—
HYPERICACEAE			
St. John's-Wort (*Hypericum perforatum* L.)	—		
IRIDACEAE			
Gladiolus (*Gladiolus* sp.)	— —		
Iris (*Iris* sp.)	—		

TABLE 7-2 Continued

Plant	Susceptible	Intermediate	Resistant
JUGLANDACEAE			
Walnut, Black (*Juglans nigra* L.)		—	
Walnut, English (*Juglans regia* L.)		—	
LEGUMINOSAE			
Alfalfa (*Medicago sativa* L.)		—	—
Bean, Snap (*Phaseolus vulgaris* L.)			—
Clover, Crimson (*Trifolium incarnatum* L.)		—	
Clover, Yellow (*Melilotus officinalis* Lam.)		—	
Locust, Black (*Robinia Pseudo-Acacia* L.)			—
Pea, Garden (*Pisum sativum* L.)			—
Soybean (*Glycine max* L.)		—	—
LILIACEAE			
Asparagus (*Asparagus* sp.)			—
Tulip (*Tulipa gesneriana* L.)	—		
MALVACEAE			
Cotton (*Gossypium hirsutum* L.)			—
MORACEAE			
Mock-Orange (*Philadelphus* sp.)			—
Mulberry, Red (*Morus rubra* L.)		—	
PINACEAE			
Arborvitae (*Thuja* sp.)			—
Fir, Douglas [*Pseudotsuga menziesii* (Mirb.) franco]	—		
Fir, Grand [Abies grandis (Dougl.) Lindl.]		—	
Juniper (*Juniperus* sp.)			—
Larch, Western (*Larix occidentalis* Nutt.)	—		

TABLE 7-2 Continued

Plant	Susceptible	Intermediate	Resistant
Pine, Eastern White, Lodgepole, Scotch, Mugo, Ponderosa (*Pinus strobus* L., *Pinus contorta* Dougl. *Pinus sylvestris* L., *Pinus mugho* Turra., *Pinus ponderosa* Laws)[b]			
(young needles)	—		
(old needles)			—
Spruce, Blue (*Picea pungens* Englm.)	—	—	
Spruce, White (young needles) [*Picea glauca* (Moench) Voss.]		—	
PLANTAGINACEAE			
Plantain (*Plantago* sp.)			—
PLATANACEAE			
Planetree, London (*Platanus acerifolia* L.)			—
Sycamore (*Platanus occidentalis* L.)			—
POLYGONACEAE			
Dock (*Rumex* sp.)			—
Smartweed (*Polygonum* sp.)	—	—	
PORTULACACEAE			
Purslane (*Portulaca oleracea* L.)			—
OLEACEAE			
Ash, Green (*Fraxinus pennsylvanica* var. *lanceolata* Borkh.)		—	
Ash, Modesto (*Fraxinus velutina* Torr.)			—
Lilac (*Syringa vulgaris* L.)		—	
Privet (*Ligustrum* sp.)			—
RANUNCULACEAE			
Peony (*Paeonia* sp.)		—	
ROSACEAE			
Apple (*Malus sylvestris* Mill.)		—	—

TABLE 7-2 Continued

Plant	Susceptible	Intermediate	Resistant
Apricot, Chinese and Royal (*Prunus armeniaca* L.)	—		
Apricot, Moorpark and Tilton (*Prunus armeniaca* L.)	—	—	
Bridalwreath (*Spiraea prunifolia* Sieb. & Zucc.)			—
Cherry, Bing and Royal Ann (*Prunus avium* L.)		—	
Cherry, Choke (*Prunus virginiana* L.)		—	
Cherry, Flowering (*Prunus serrata* L.)		—	—
Firethorn (*Pyracantha* sp.)			—
Mountain Ash, European (*Sorbus aucuparia* L.)		—	
Peach (fruit) (*Prunus persica* Sieb. & Zucc.)	—		
Peach (foliage) (*Prunus persica* Sieb. & Zucc.)		—	
Pear (*Pyrus communis* L.)			—
Plum, Bradshaw (*Prunus domestica* L.)	—		
Plum, Flowering (*Prunus cerasifera* Enrh.)		—	—
Prune, Italian (*Prunus domestica* L.)	—		
Rose (*Rosa odorata* Sweet.)		—	
Raspberry (*Rubus idaeus* L.)		—	—
Serviceberry (*Amelanchier* sp.)	—	—	
Strawberry (*Fragaria* sp.)		—	—
RUBIACEAE			
Coffee (*Coffea arabica* L.)			—
RUTACEAE			
Grapefruit (*Citrus paradisi* Mact.)		—	
Lemon (*Citrus* sp.)		—	
Orange (*Citrus sinensis* Osbeck.)		—	

TABLE 7-2 Continued

Plant	Susceptible	Intermediate	Resistant
Tangerine (*Citrus* sp.)		—	
SALICACEAE			
Aspen, Trembling (*Populus tremuloides* Michx.)		—	
Poplar, Balsam (*Populus balsamifera* L.)			—
Poplar, Carolina (*Populus eugenei* Simon-Louis)		—	
Poplar, Lombardy (*Populus nigra* L.)		—	
Willow (*Salix* sp.)			—
SAXIFRAGACEAE			
Currant (*Ribes* sp.)			—
SIMAROUBACEAE			
Tree of Heaven (*Ailanthus altissima* L.)			—
SOLANACEAE			
Eggplant (*Solanum melongena* L.)			—
Jerusalem Cherry (*Solanum Pseudo-Capsicum* L.)	—		
Nightshade (*Solanum nigrum* L.)			—
Pepper, Bell (*Capsicum frutescens* L.)		—	—
Petunia (*Petunia* sp.)			—
Potato (*Solanum tuberosum* L.)			—
Tobacco (*Nicotiana tabacum* L.)			—
Tomato (*Lycopersicon esculentum* Mill.)		—	—
TAXACEAE			
Yew, Spreading (*Taxus cuspidata* Sieb. & Zucc.)		—	
THEACEAE			
Camellia (*Camellia japonica* L.)			—

TABLE 7-2 Continued

Plant	Susceptible	Intermediate	Resistant
TILIACEAE			
Linden, American (*Tilia americana* L.)			—
Linden, European (*Tilia cordata* Mill.)		—	
ULMACEAE			
Elm, American (*Ulmus americana* L.)			—
Elm, Chinese (*Ulmus pumila* L.)			—
UMBELLIFERAE			
Carrot (*Daucus carota* L.)			—
Celery (*Apium graveolens* L.)			—
VIOLACEAE			
Violet (*Viola* sp.)		—	
VITACEAE			
Grape, Concord (*Vitis labrusca* L.)		—	
Grape, European (*Vitis vinifera* L.)	—		
Virginia Creeper (*Parthenocissus quinquefolia* Planch.)			—

[a] The rating of a plant with respect to its susceptibility to hydrogen fluoride is somewhat subjective and can be modified by environmental and biologic factors, as discussed in the text.

[b] Some populations may exhibit a wide range of tolerances to fluoride.

genus with respect to susceptibility is shown by the statement of Ender[191] that *Larix decidua* and *L. leptolepsis* are resistant, whereas Guderian *et al.*[239] show *L. leptolepsis* as highly susceptible.

- Most of the broad-leaved deciduous trees of importance to forestry are of intermediate susceptibility, or relatively tolerant, to fluoride symptoms.
- Most deciduous trees and shrubs important as ornamentals and floricultural crops are of intermediate susceptibility or are

tolerant. Some maples may be susceptible enough to be considered exceptions.

- Common vegetable crops and field crops, including forage, are tolerant or only moderately susceptible. Sweet corn, sorghum, and possibly a few other grasses are notable exceptions, and many varieties must be considered susceptible. Only Guderian *et al.*[239] list a number of legumes, placing most of them in the most susceptible class. This is not consistent with the listing by other authorities or with the general statement that most forage crops are not highly susceptible.[93] This is the result of the classification system used by Guderian *et al.*[239] Their list of agricultural crops contains only legumes, grasses, some mustards, and a few other species generally considered of intermediate susceptibility or resistant. Their highly susceptible class in this group indicates only that these are more susceptible than others within the group. They do not intend to suggest that the clovers and alfalfa listed above have the same susceptibility as, for example, the members of the lily family, which are classified as ornamentals. The grasses are mostly intermediate in susceptibility. Only one species of grass, wheat, is included in their least susceptible category.
- Practically all important fruits and berries must be considered at least moderately susceptible. Some species and varieties are very susceptible: peaches, some apricots, blueberries, and some grapes. The apricot is a classic example of varietal differences in susceptibility. The Chinese varieties are highly susceptible, but the Moorpark variety is more resistant. The hybrid roses, although generally considered of intermediate tolerance to fluoride, also exhibit considerable variation in susceptibility among varieties.[107]
- The nonwoody flowering ornamentals and floricultural crops are a heterogeneous group. Most, if not all, spring-flowering bulbs are highly susceptible. Many varieties of gladiolus, important as ornamentals and as floricultural crops, are among the most susceptible plants known. Members of the pink and composite families are generally relatively tolerant.

PLANT RESPONSE IN RELATION TO AIR QUALITY

McCune[424,425] examined the available literature up to 1968 reporting exposure of plants to fluoride in which the concentration of gaseous fluoride and the duration of exposure could be definitely established.

His report presented plots of the available data for eight groups of plants: tomato, alfalfa, sorghum, corn, gladiolus, citrus, other tree fruits, and conifers. The plots consist of a notation of response (foliar symptoms, no symptoms, yield effect, and so on), in a field defined by the log of the mean atmospheric concentration (in micrograms of fluoride per cubic meter), and of exposure for 1–20 days. The only significant publication not included is that of Guderian *et al.*[239]

The plots from McCune's papers are reproduced as Figures 7-1 through 7-13 with the following modifications: the alfalfa data have been expanded, data on other forage crops have been compiled, and the data of Guderian *et al.*[239] have been included. Although the data in the figures refer to many types of responses, this discussion considers each effect separately.

Foliar Lesions

There are two practical aspects to the presence of foliar lesions on plants. If the aesthetic quality of a plant depends on the appearance

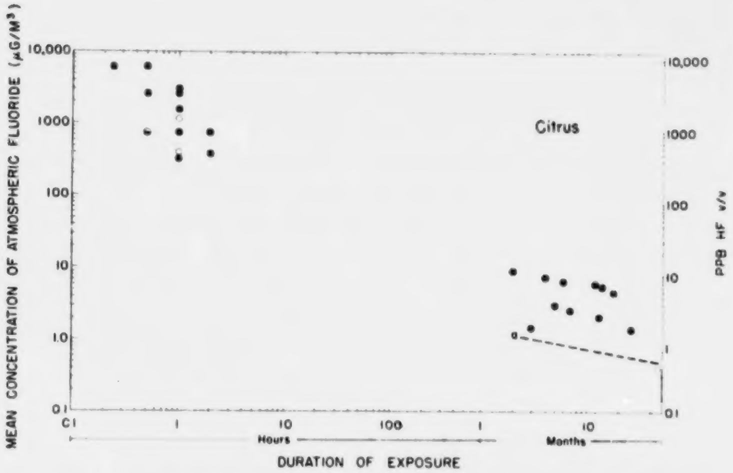

FIGURE 7-1 Relation of concentration and duration of exposure to effects of atmospheric fluoride on citrus. ○, no foliar markings observed; ●, markings present; ◒, threshold or point at which markings first appear. (Reprinted with permission from McCune.[425])

of its foliage, necrotic or chlorotic lesions will have an obvious detrimental effect. However, if the yield of the plant is important, foliar lesions may be indirectly related to an effect on yield. Hill[275] has suggested that necrosis of 5% of the foliar mass might be viewed as the maximal permissible effect. In gladiolus, significant effects on corm and flower yield have been demonstrated when there was 10–70% leaf necrosis,[103,239] or when more than 10% of the leaf mass was removed.[149]

CITRUS

Because of its growth habit of periodic leaf flushes and foliar longevity, citrus is unique among economically important fruit. The limited data available are presented in Figure 7-1. McCune suggests the line *a* as a reasonable criterion for citrus exposure to fluoride, fully recognizing that the data are very limited. Both leaf symptoms and growth effects have been reported.[101,108,396,675] The more recent data of Brewer *et al.*[105] are not included in this summary but are consistent with the suggestions made by McCune.

TREE FRUITS (EXCLUDING CITRUS)

McCune[425] grouped all the deciduous fruit trees in Figure 7-2:

> This category of plants will be subject to the influence of atmospheric fluorine during a limited part of the year, the growing season from bud break to leaf fall, and in this respect it is similar to previous examples. Unlike the species discussed earlier, it is a perennial plant, but, unfortunately, data are not available for effects in successive years, or indeed for periods over 40 days. . . . The only response considered is the presence of foliar markings[8,68,164,165,238,276,617,669,741,742] and its relationship to growth or yield is unknown. The appearance of chlorosis or necrosis on leaves has not been reported for time–concentration values lying below line *a* and both the presence and absence of markings are found between lines *a* and *b*. It is not possible to establish a reasonable criterion for long-term effects on growth or yield.
>
> The effects on apples, cherries, peaches, prunes, and apricots are considered together due to the limited amount of data on any one of these species. Specific and varietal differences in susceptibility are known to be present and contribute to the heterogeneity. However, the relative susceptibility of a fruit crop may vary with concentration of atmospheric fluorine.[8] The establishment of a criterion for the entire group may be justified if it is common for more than one type of fruit crop to be present in a given locality.

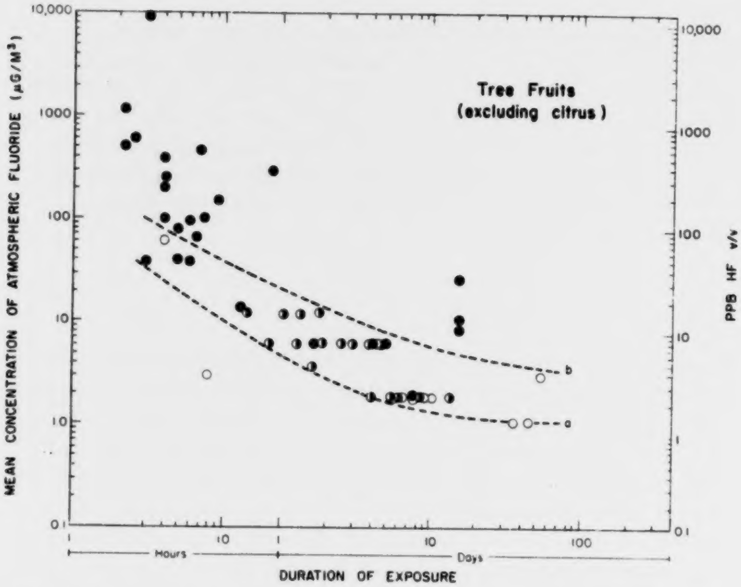

FIGURE 7-2 Relation of concentration and duration of exposure to effects of atmospheric fluoride on tree fruits, excluding citrus. ○, no foliar markings observed; ○, markings present; ◑, threshold or point at which markings first appear. (Reprinted with permission from McCune.[425])

CONIFERS

All the conifers for which data are available are grouped in Figure 7-3. These data are only in reference to appearance of necrosis on needles. The discussion presented by McCune[425] follows:

> Differences in generic, specific, and individual susceptibility account for some of the heterogeneity shown by the data. The points at which trace amounts of necrosis could occur lie along line *a* for larch and line *b* for western white pine, lodgepole pine, arbor vitae and spruce. Perhaps the more important factor in considering susceptibility to fluoride is the stage of development of the needles. The points at which traces of injury would appear on ponderosa pine are indicated by line *c* for needles that have elongated for two to three months and by line *d* for needles that are about one month less mature.[15] Needles that have elongated in the previous years might be more resistant than those represented by line *c*.

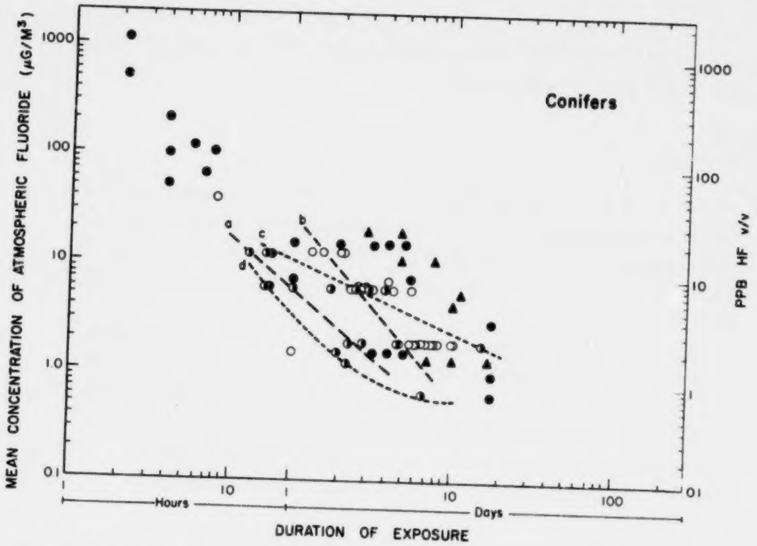

FIGURE 7-3 Relation of concentration and duration of exposure to effects of atmospheric fluoride on conifers. ○, no foliar markings observed; ●, markings present; ◑, threshold or point at which markings first appear. Curves represent possible thresholds for slight needle injury; a, larch; b, western white pine, lodgepole pine, arborvitae, and spruce; c, mature needles of ponderosa pine; and d, young needles of ponderosa pine, (Reprinted with permission from McCune.[425])

No data are available that can directly relate time–concentration values or injury on needles to effects on growth or productivity. Moreover, there is a lack of data on dose–response relationships for periods over a month. On the basis of our present knowledge, perhaps a suitable criterion would be a line, such as *d*, that is based on injury to the young needles during their period of expansion.

The points supplied by Guderian *et al.*[239] for needles of white pine of different ages agree with results of others and would fall on or near line *d*.

Growth and Reproduction

For most plants, the commercial value and interest in the effects of fluoride lie in the yield of the crop, which is determined by the growth and reproduction of the plant. Information on growth and reproduction is not as voluminous as that on foliar lesions. Growth and reproduction, as well as foliar lesions, of some groups of plants are considered in this section.

TOMATO

Figure 7-4 shows the available data on tomato. Lines *a* and *b* are the suggested criteria for effects, as given by McCune. Line *a* is suggested as a reasonable limit of foliar effects, and line *b* as the limit for economic effects, such as reduction in quantity or quality of the crop.

The problems associated with attempting to draw boundaries between response and no response (in relation to exposure) are even more difficult than those associated with relating accumulation to exposure or response to accumulation.

Figure 7-4 shows no clear-cut boundary between presence and absence of foliar markings. The original authors may have termed

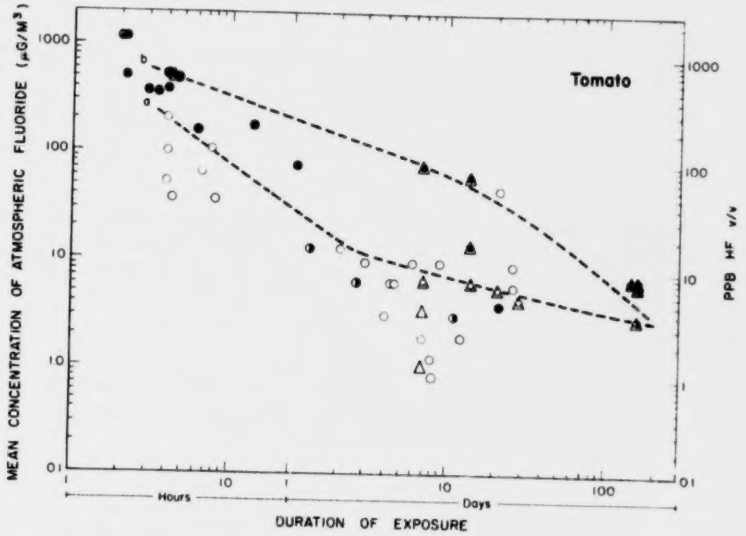

FIGURE 7-4 Relation of concentration and duration of exposure to effects of atmospheric fluoride on tomato. ○, no foliar markings observed; ●, markings present; ◑, threshold or point at which markings first appear; Δ, no effect on yield; ▲, significant reduction in yield. (Reprinted with permission from McCune.[425])

the effect severe, moderate, or trace, but there is no good way to collate the results from several investigators. Environmental factors and stage of growth are additional variables that are difficult to assess. Zimmerman and Hitchcock[742] have demonstrated that plants grown with adequate soil moisture will develop foliar lesions after a given exposure to hydrogen fluoride, but plants grown with restricted soil moisture will develop no markings. Tomatoes grown in conditions of optimal nitrogen, phosphorus, and calcium were more severely marked than those grown under nutritional conditions above or below the optimum for "best" growth.[98] Other work has suggested that reduced calcium content increased the susceptibility of tomato to leaf markings, as well as to effects on yield as a result of exposure to fluoride.[503] Line *a* in Figure 7-4 represents a considered judgement of the available data. Line *b*, as noted by McCune, "represents a boundary of present knowledge and may not be suitable [as a criterion] if the data on yield effects are considered too limited."

CORN

Figure 7-5 presents data for corn. The short line *a* is suggested as the boundary between presence and absence of detectable chlorosis on the susceptible varieties. Line *b* represents the same boundary for the resistant varieties. Line *c* is suggested as a possible threshold for growth effects observed when the more susceptible varieties are exposed during tassel emergence. This stage is the period of greatest susceptibility of corn to effects on growth and yield. McCune comments:[425]

> A line, such as *d*, would appear to be a reasonable criterion for effects on corn within the limited scope of the data. The data on fluorine accumulation are insufficient to determine its relationship to air quality. The grain shows no significant accumulation of fluorine[280] but fluorine accumulation by the foliage may be important when the whole plant is used for ensilage. A different criterion, namely one based on analysis of plant tissue, will have to be used as is the case with forage.

SORGHUM

Figure 7-6 presents data for sorghum. McCune suggests lines *a* and *b* as reasonable boundaries for the appearance of chlorosis in susceptible varieties (line *a*) and the more resistant varieties (line *b*). Growth and yield of the more susceptible varieties are affected at exposures greater

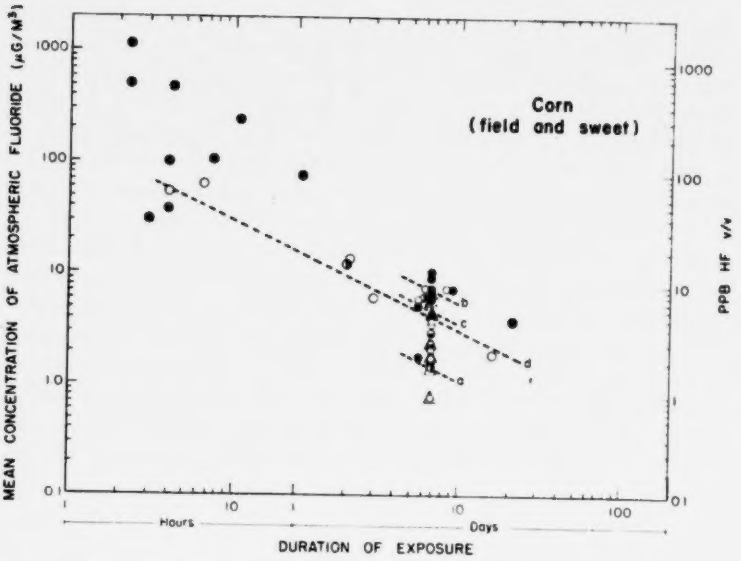

FIGURE 7-5 Relation of concentration and duration of exposure to effects of atmospheric fluoride on corn. ○, no foliar markings observed; ●, markings present; ◑, threshold or point at which markings first appear; Δ, no significant effect on yield. (Reprinted with permission from McCune.[425])

than represented by line *c*, and in the more resistant varieties at exposures above those represented by line *d*. McCune continues:[425]

> Significant effects on growth and yield were commonly associated with pronounced foliar chlorosis; however, in one case a 20 per cent reduction in yield was found in the absence of chlorosis. Effects on growth were usually associated with effects on yield although one could be present in the absence of the other. One reason for these anomalies is the effect of developmental stage on the susceptibility of sorghum to atmospheric fluorine. Plants were more sensitive to effects on yield at the early heading stage. Moreover, exposure to atmospheric fluorine at this stage could result in effects on flowering and yield that would not be closely associated with chlorosis or reduced growth. The criterion represented by line *c* would probably lie next to line *b* if this critical stage were not considered.
>
> A line, such as *c*, would probably be a reasonable criterion for this crop considering the effects of variety and stage of development on its sensitivity to fluorine. Although there are no data available for exposure periods of a month or growing season, a criterion based upon periods of exposure up to three weeks may be sufficient if established with reference to a period of maximum sensitivity or critical importance to the life and use of the plant. The one- to two-week

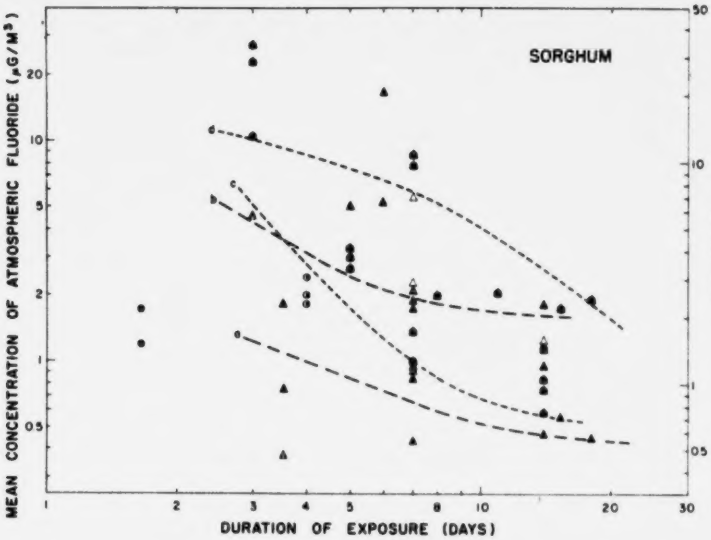

FIGURE 7-6 Relation of concentration and duration of exposure to effects of atmospheric fluoride on sorghum (Milo maize). ○; no foliar markings observed; ●, foliar markings present; ◐, threshold; Δ, no effect on growth or yield; ▲, significant reduction in growth or yield. (Reprinted with permission from McCune.[425])

period when the head is emerging is regarded as the period of greatest susceptibility for the plant, and the data used in deriving line *c* are based upon effects of atmospheric fluorine during this stage of development. This criterion is also based on the effects of fluorine on the variety of greater susceptibility since it is assumed that a variety would be chosen for its suitability to a particular agronomic situation and not for its resistance to fluorine. This criterion would allow a certain amount of chlorosis but it should be protective of yield or growth.

GLADIOLUS

Data on gladiolus are extensive, and Figure 7-7 presents the assessment made by McCune.[425] Considering the varietal differences, as well as differences in experimental procedures and environmental conditions represented in these data, agreement is quite good.

As Hitchcock *et al.*[281] have noted, it is often difficult to find gladiolus without some slight tip burn, even with no apparent source of atmospheric fluoride. The only data relating effects on flower or corm production to time and concentration values are from these authors.[281]

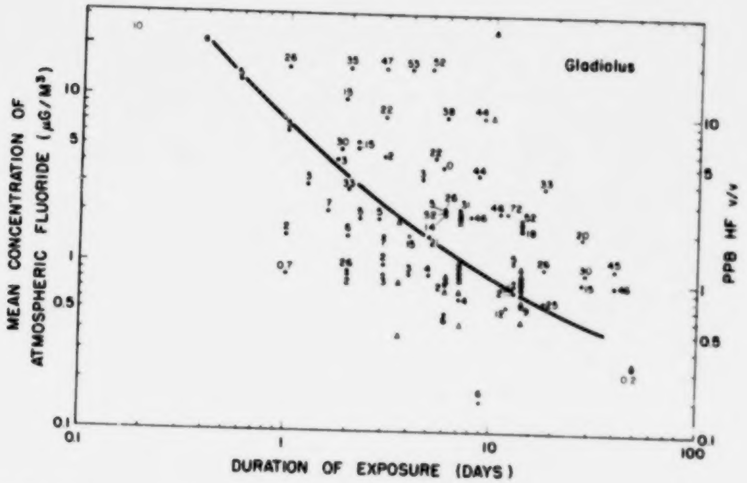

FIGURE 7-7 Relation of concentration and duration of exposure to effects of atmospheric fluoride on gladiolus. The degree of leaf-area necrosis, in percent, induced by atmospheric fluoride is shown. In addition, fluoride-induced effects on flowering or corm production are shown: Δ, no effect; ▲, significant reduction observed. (Reprinted with permission from McCune.[425])

McCune suggests line *a* as the boundary for about 5% tip burn, in accordance with the relation between tip burn and yield discussed previously.

Accumulation of Fluoride by Plants

With other types of crops, the accumulation of fluoride by the plant could be incidental to its effects on the cellular, foliar, or organismic level. But with alfalfa or other forage crops, the accumulation of fluoride is important.

Unfortunately, the information available is limited, for technical and other reasons, to exposure periods that do not reflect the temporal distribution of atmospheric fluorides and the influence of climatic factors during exposures that would be encountered in the field situation. Moreover, a limited range of species and varieties have been tested and there are difficulties inherent in comparing results between laboratories that use different equipment, methods of analysis or exposure techniques. With these qualifications in mind, the data presented here on the responses of alfalfa and orchard grass may be considered as illustrating some aspects of the relation of air quality to fluoride accumulation, as well as to foliar lesions and growth in forage crops.

ALFALFA

Figure 7-8 shows the results of about 300 fumigations of alfalfa in field plots over a 13-year period[279] (circles) and recent data published by Treshow and Harner[680] and Guderian *et al.*[239] (triangles) superimposed over the older results published by McCune[425] (inverted triangles). All the points shown refer to the presence (filled symbols) or absence (open symbols) of foliar lesions. There is no definite boundary for the occurrence of foliar lesions, in that seedlings may be more susceptible than more mature plantings and the rate of air flow past the plant can alter susceptibility. Thus, a reasonable boundary for the presence of foliar lesions in seedling alfalfa may be represented by curve *a*, and in mature plants by curve *b*.

Although foliar lesions are the most easily and extensively documented responses of plants to fluoride, an effect on growth, as presented in Figure 7-9, would be more relevant to alfalfa (the symbols

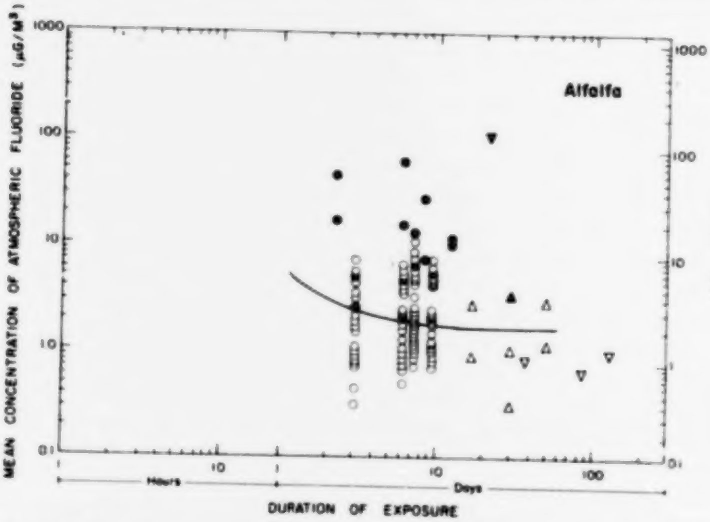

FIGURE 7-8 Relation of concentration of hydrogen fluoride and duration of exposure to growth effects in alfalfa. (See text for explanation of symbols.)[426]

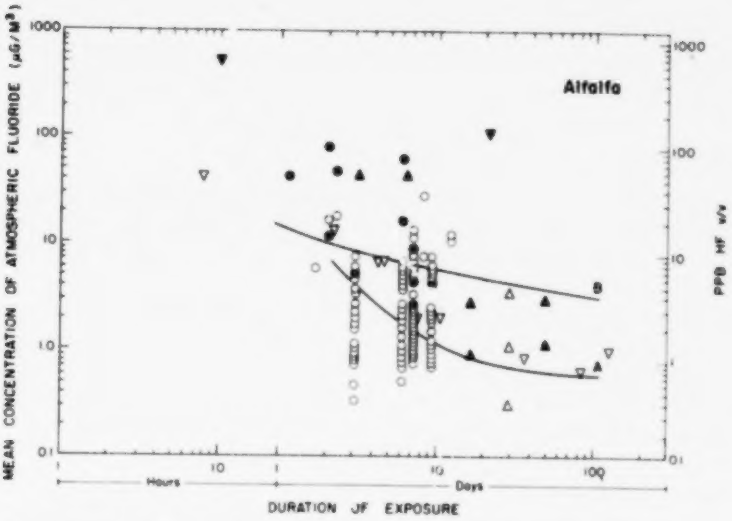

FIGURE 7-9 Relation of concentration of hydrogen fluoride and duration of exposure to the production of foliar lesions on alfalfa. (See text for explanation of symbols.) (Reprinted with permission from McCune.[426])

are the same as those used in Figure 7-8). The only growth effects reported by Hitchcock *et al.*[279] constitute an increased fresh weight of the plant, but those found by other investigators[67,680] constitute a decrease in the dry weight of the plant. Treshow and Harner[680] reported that a decrease in dry weight occurred in the absence of foliar lesions, but Guderian *et al.*[239] found no significant effect on yield in plants showing slight amounts of chlorosis or necrosis. A line designating the threshold for effects on growth may be that shown in Figure 7-9.

Effects of fluoride on the flowering of alfalfa were also investigated, but none that could be considered significant was found.[279]

The best estimate of the relation of the concentration of fluoride in alfalfa to concentration of hydrogen fluoride and duration of exposure was given by the linear regression equation, $F = 1.89CT + 0.74$, in which F is fluoride concentration, in ppm; C is concentration of hydrogen fluoride, in μg of fluoride per cubic meter; and T is duration of exposure, in days.[279,428] This value, obtained from exposure periods of 9 days or less, is relatively close to the value of 2.4 computed for alfalfa on the basis of other results[66,67,239] for periods greater than 16 days.

However, neither of these values for the "accumulation coefficient" takes into account the decrease in the fluoride concentration observed in the postexposure period. The estimated half-life of the fluoride concentration was found to lie between 8 and 22 days, and the data were reanalyzed and tested for goodness of fit to a second model, which assumed that uptake of fluoride was directly proportional to the product of time and concentration and that loss of fluoride was a first-order reaction.[279,428] The equation, $F = 4.22\, C^{0.82}\, T^{0.70}\, e^{-0.032D}$, resulted, in which F is fluoride concentration in alfalfa, in ppm; C is concentration of hydrogen fluoride, in μg of fluoride per cubic meter; T is duration of exposure, in days; and D is length of postexposure period, in days. The curves shown in Figure 7-10 illustrate the relation between fluoride accumulation in alfalfa and duration of exposure and concentration of hydrogen fluoride with no postexposure period. The curves shown in Figure 7-11 illustrate the same relation with various postexposure periods and a constant fluoride concentration of 40 ppm in alfalfa. There are three possible causes for the decrease in the fluoride concentration of alfalfa with time: dilution by growth after fumigation; loss of older, fluoride-containing leaves; and loss of

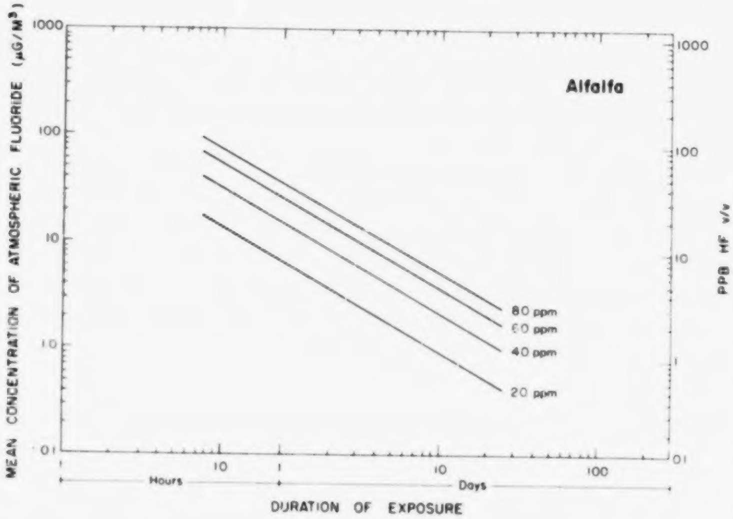

FIGURE 7-10 Relation of concentration of hydrogen fluoride and duration of exposure to accumulation of fluoride in alfalfa. (Reprinted with permission from McCune.[426])

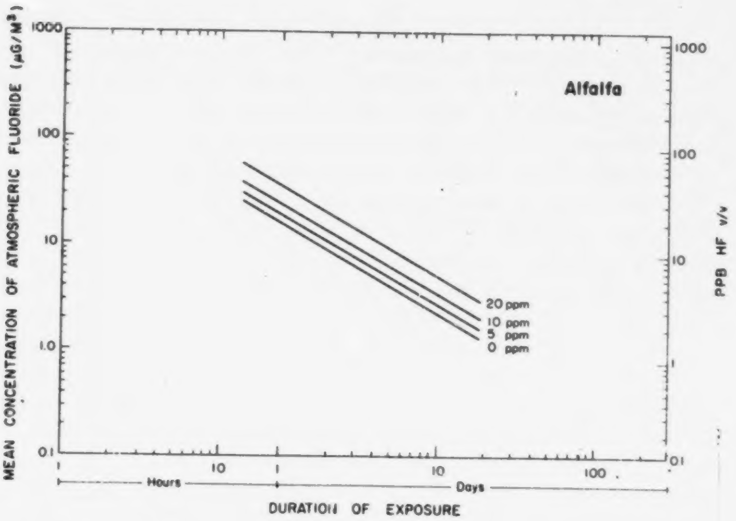

FIGURE 7-11 Relation of concentration of hydrogen fluoride and duration of exposure to give an accumulation of 40 ppm of fluoride in alfalfa subjected to different postexposure periods. (Reprinted with permission from McCune.[426])

fluoride from the leaves by weathering. However, the relative effectiveness of each of these in lowering the fluoride concentration is not known.

The data for fumigation studies with alfalfa reported by Benedict *et al.*[66,67] and Guderian *et al.*[239] are summarized in Table 7-3. The K values were calculated from the data given by substituting in the equation $\Delta F = KCT$, in which ΔF is in parts per million of fluoride on a dry-weight basis (difference between unwashed fumigated and control samples). Time is measured in days, and concentration is expressed in micrograms of fluoride per cubic meter of air.

ORCHARD GRASS

In an analysis of data on the accumulation of fluoride by orchard grass, Hitchcock *et al.*[279,428] reported that the stepwise regression of accumulated fluoride on first- and second-degree terms of concentration and time gave the equation $F = 1.13CT - 1.17$, in which F is concentration in orchard grass, in ppm; C is mean concentration

TABLE 7-3 Fluoride Uptake by Alfalfa

Fumigation concentration of fluoride, $\mu g/m^3$	Duration of fumigation, days	Fluoride content, ppm (dry weight)		K, $\Delta F/(\mu g/m^3)(days)$	Reference
		Fumigated	Control		
0.6	80	75	10	1.4	67
0.7	23	20	14	0.4	67
0.7	45	80	14	2.1	67
0.7	78	102	14	1.6	67
0.8	35	124	6	4.2	67
0.8	60	156	(10)[a]	3.0	66
0.9	120	178	6	1.6	67
0.85	16	51	20	2.3	239
1.1	47.7	145	40	2.0	239
2.6	16.0	150	20	3.1	239
2.8	47.7	411	40	2.8	239
5.0	1	37	(10)[a]	5.4	66
5.0	2.3	74	(10)[a]	7.7	66
40	3.125	803	36	6.1	239
40	6.25	1327	36	5.2	239

[a] Not stated in original paper; 10 ppm assumed from similar data in Benedict *et al.*[67]

of hydrogen fluoride, in μg of fluoride per cubic meter; and T is the duration of exposure, in days. Figure 7-12 shows the intersections of the regression surface with planes representing 20, 40, 60, and 80 ppm, projected onto the time–concentration plane. No significant effect was found for the duration of the postexposure period for orchard grass.

The data on orchard grass reported by Benedict *et al.*[66,67] and Guderian *et al.*[239] are summarized in Table 7-4.

OTHER FORAGE CROPS

Additional data for other forage crops from the report by Guderian *et al.*[239] are summarized in Table 7-5. A variety of crops were exposed under comparable conditions for 16 days (384 hr) at fluoride concentrations of 0.85 and 2.6 $\mu g/m^3$ and for 47.7 days (1145 hr) at 1.1 and 2.8 $\mu g/m^3$. Table 7-5 gives the K values calculated from the original data. There is no consistent dependence on time or concentration in this limited study. The average K value is calculated and presented with its reciprocal.

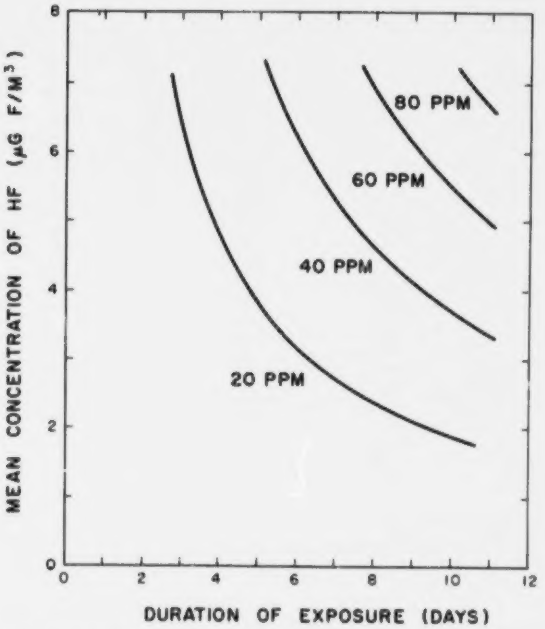

FIGURE 7-12 Relation of fluoride concentration in orchard grass to concentration of hydrogen fluoride and duration of exposure. (Reprinted with permission from Hitchcock *et al.*[279])

DISCUSSION

It is apparent, from the range of values of accumulation coefficients (K) for alfalfa, orchard grass, and other forage crops, that a difference in susceptibility to fluoride among species, or even varieties, is to be expected.

The data of Benedict *et al.*[66,67] and Guderian *et al.*[239] suggest that the K values may be proportional to air concentration, particularly in the case of alfalfa. If that is true, there is no obvious physical or biologic explanation. However, the exponential coefficient for concentration in the equation supplied by Hitchcock *et al.*[279,428] indicates that the apparent accumulation coefficient decreases with increasing concentrations.

The data for both alfalfa and orchard grass suggest that the K values are inversely proportional to time. The exponential coefficient for

TABLE 7-4 Fluoride Uptake by Orchard Grass

Fumigation concentration of fluoride, $\mu g/m^3$	Duration of fumigation, days	Fluoride content, ppm (dry weight)		K, $\Delta F/(\mu g/m^3)(days)$	Reference
		Fumigated	Control		
0.6	80	40.0	6.0	0.7	67
0.7	23	12.0	5.2	0.4	67
0.7	45	32.0	5.2	0.8	67
0.7	78	41.0	5.2	0.7	67
0.8	21	35.4	5.5	1.8	67
0.8	35	45.0	14.4	1.1	67
0.8	60	172	(6)[a]	3.5	66
0.9	120	229	6	2.1	67
0.85	16	41	11	2.2	239
1.1	47.7	137	22	2.2	239
2.6	16	98	11	2.1	239
2.8	47.7	282	22	2.0	239
40	3.125	496	19	3.8	239

[a] Not stated in original paper; 6 ppm assumed from similar data in Benedict *et al.*[67]

time, in the equation for alfalfa, also indicates that the apparent accumulation coefficient decreases with increasing periods of exposure.[279,428] Three possible factors could account for this relation:

A saturation effect may be operative, tending to reduce absorption rate as fluoride builds up in the tissue

Changes in the plant tissues as they mature may reduce absorption rate

The mass of the plant at the beginning of the fumigation period, in relation to the mass added during fumigation, could affect the apparent accumulation coefficient

As a plant matures, total surface area and mass approach limiting values. That fact would work against the observed decrease of *K* with time. Regardless of the reasons, the relation seems, on the basis of the available data, to be real.

The average exposure time represented in Tables 7-3 and 7-4 is about 40 days. The average *K* for periods of 16 days or more, except for the anomalous values (23 days as in line 2 of Tables 7-3 and 7-4 and 60 days as in line 7 of Table 7-4), is 2.4 for alfalfa and 1.4 for orchard grass.

In field problems, we are generally concerned with 30–60 days of exposure, and for many purposes we would be interested in fluoride

TABLE 7-5 Coefficient of Accumulation, *K*, for Various Forage Crops[a]

	Fluoride Concentration and Duration of Exposure					
Crop	0.85 $\mu g/m^3$, 16 days	2.6 $\mu g/m^3$, 16 days	1.1 $\mu g/m^3$, 47.7 days	2.8 $\mu g/m^3$, 47.7 days	Average	1/*K* (avg.)
Italian rye grass (*Lolium multiflorum*)	4.3	6.3	2.3	4.1	4.25	0.24
Perennial rye grass (*Lolium perenne*)	5.1	3.1	4.1	3.0	3.82	0.26
Orchard grass (*Dactylis glomerata*)	2.2	2.1	2.2	2.0	2.12	0.47
Fescue (meadow) (*Festuca pratensis*)	3.4	4.7	3.0	3.5	4.87	0.20
Timothy (*Phleum pratense*)	1.3	2.2	2.2	3.7	2.35	0.42
Alfalfa (*Medicago sativa*)	2.3	3.1	2.0	2.8	2.55	0.39
Red clover (*Trifolium pratense*)	2.0	4.1	3.8	5.4	3.82	0.26
White clover (*Trifolium repens*)	4.3	4.8	5.9	9.4	6.10	0.16
Alsike clover (*Trifolium hybridum*)	3.5	3.9	–	–	3.70	0.27
Crimson clover (*Trifolium incarnatum*)	3.4	4.7	–	–	4.05	0.25

[a] Adapted from Guderian *et al.*[339]

accumulations of 30–40 ppm in tissue. Therefore, the problem is to relate air quality to the accumulation of these levels of fluoride and these periods of exposure. Three approaches to this problem are suggested.

If we use the equation, $\Delta F = KCT$, and assume ΔF to be 40 ppm and T to be 60 days, with K equal to 1.4, the estimated values of C would be equal to about 0.5 $\mu g/m^3$ for orchard grass. For alfalfa, with K equal to 2.4, the value of C would be about 0.3 $\mu g/m^3$. Of course, this approach ignores two factors: the decrease in the apparent accumulation coefficient and the decrease in the fluoride concentration of the plant with time. Additionally, the standard deviations from regression reported by McCune and Hitchcock[428] of 13.8 ppm and 16.8 ppm for orchard grass and alfalfa, respectively, indicate that point estimates based on their regression equations would have relatively broad confidence limits.

The effects of these two factors can be illustrated by using the equation of Hitchcock *et al.*[279,428] for alfalfa, with the assumption that the crop will have a concentration of fluoride equal to 40 ppm at the end of a 60-day period. A 10-day period of exposure to hydrogen fluoride beginning the 60-day period must average 15.3 $\mu g/m^3$, giving a mean for the 60-day period of 2.6 $\mu g/m^3$. But a 10-day period of exposure ending the 60-day period must average 2.2 $\mu g/m^3$, giving a mean of 0.4 $\mu g/m^3$ for the entire 60-day period. Thus, the effect of time on the accumulation coefficient and the duration of the post-exposure period indicate the importance of the sequence of exposures in determining the relation of air quality to fluoride accumulation.

The experimental data summarized here cover two different situations. Estimates of fluoride accumulation coefficients are based, on the one hand, on long-term fumigations without postfumigation exposures to weathering,[66,67,239] and, on the other hand, on short-term fumigation followed by postfumigation periods of 3–15 days in ambient field conditions.[279] The consistency of the data obtained from three widely different environmental regions–New York, California, and Germany–with different cultural characteristics suggests that the uncertainty may be less than a factor of 2.

It is unfortunate that there are no data available from ambient air situations that relate fluoride accumulation to air quality, especially because in ambient conditions plants are exposed to both gaseous and particulate fluoride compounds and to continuous weathering and other environmental factors.

SUMMARY

Figure 7-13 summarizes published suggestions[425] for criteria of plant response to various concentrations of atmospheric fluoride for different plant species. For the three most susceptible groups considered–gladiolus, sorghum, and conifers– the suggested criteria lines terminate at 0.5 $\mu g/m^3$. The citrus data suggest about 0.5 $\mu g/m^3$ for a long-

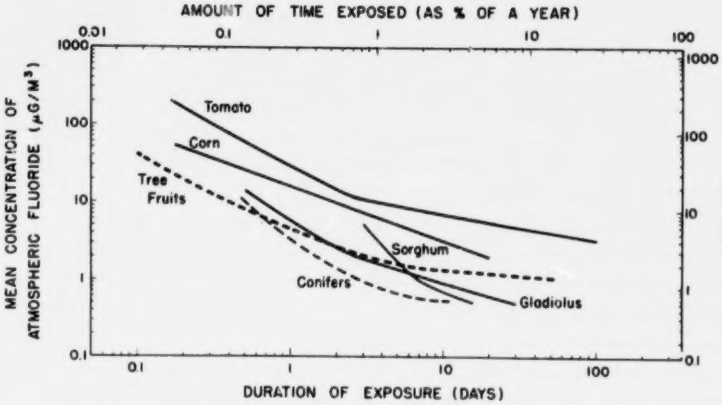

FIGURE 7-13 Possible air-quality criteria for atmospheric fluoride, with reference to different plant species. (Reprinted with permission from McCune.[425])

term exposure limit for a significant effect. Within a factor of 2, this concentration of fluoride might hold for fluoride accumulation in forage crops.

Describing the relation of air quality to effects on vegetation involves both uncertainty and complexity. One reason for the complexity is that a plant's response to fluoride is influenced by many different factors. The existence of many environmental (climatic and edaphic) factors implies that the effect of atmospheric fluoride may vary with the site or the agricultural practices used, and that, if the conditions in which the experimental data were obtained differ from those in which the data will be used, an extrapolation from laboratory effect to field event is necessary.

The biologic factors also have implications. Different species of plants or even different varieties of the same species will differ in their response or susceptibility to fluoride, and the response of a given plant may not be the same at different stages in its life cycle.

The specific pollutant and type of exposure constitute another influence. The temporal pattern of exposure may be as important as the duration of the exposure or the mean concentration of fluoride in determining the effect of fluoride on a plant. A plant may also be expected to respond to "air pollution," as well as to a single pollutant.

But there are no adequate data on the interactive effects on the plant of fluoride and other pollutants.

Another reason for the complex nature of a plant's response to fluoride is the diversity of the effects themselves. One can classify the botanic effects of fluoride with respect to the level of biologic organization: metabolic and cellular disruption; lesions of tissues or organs; altered growth or reproduction of an entire plant; and alterations in the ecosystem. Unfortunately, the connection of an effect on one level with that on another is difficult at best and is possible in only a few cases. For example, it is possible to correlate percentage of necrotic tissue with yield of corms in gladiolus or fluoride accumulation with lesions for some plants.

But the interpretation of fluoride-induced effects is also important and complex. The effects found in scientific investigations must be judged in relation to the ecologic, aesthetic, or commercial role of a plant. One would consider death or discoloration of the foliage of ornamental plantings around a residence to be an example of aesthetic damage. The commercial value of a plant would be damaged by a reduced quantity or quality of the product. The quality of the plant product could be damaged by an effect on the external condition or appearance, such as discoloration of fruit, or an effect on the internal components, such as sugars, acids, and proteins. A change in an internal component would constitute damage if it decreased the value as a raw material, such as the sugar in sugar beets or cane, or the value as a biologically important substance, such as the nutritive value of a food or forage crop. To this last type of effect should be added the accumulation of fluoride above permissible concentrations in food products.

The available information is insufficient in depth and scope to allow unequivocal statements to be made about the effects on plants of fluoride at low atmospheric concentrations. One reason for the lack of information is the paucity of experiments designed to relate air quality to effects on plants. A second is the lack of sufficient ambient-air monitoring in connection with field studies and surveys, due in part to the lack of accurate and precise methods for the separation and collection of particulate and gaseous fluorine compounds. A third reason is the inadequacy of present experimental techniques for long-term studies in which field conditions can be simulated.

The way in which one uses and extrapolates information on the relation of atmospheric fluoride and plant response will depend on the answers to several questions: What is the most important effect that should be considered? What is the most significant segment of vegetation to be protected? What range of environmental conditions will be of concern? What are the temporal factors in terms of duration of exposure and stage of plant development? The answers to these questions must be based on the objectives of air quality. In the simplest terms, the minimal effective concentrations of atmospheric fluoride would produce either no necrosis or an acceptable amount of necrosis on susceptible receptor plants or would result in a tolerable accumulation of fluoride in forage.

8

Effects of Fluoride on Animals

Damage to cattle, sheep, horses, bees, and various species of wildlife has been claimed in areas in which fluoride is a known industrial pollutant. Present knowledge of the effects of fluorides on species of livestock that are affected by fluoride as an industrial pollutant has been gained both from observations of cases of fluoride toxicity in the field and from experimental studies. Although other studies will be referred to for specific points, most of the published work on experimental fluoride toxicity has been carried out by three research groups in the United States. A group at the University of Tennessee has studied the effects of fluoride on beef cattle, and their work is available largely in the form of University of Tennessee technical bulletins;[283,284] the original data from recent experiments at Utah State University on the effects of sodium fluoride on dairy cattle were published in a series of papers[261,587,588,632,633] and have been summarized in another report;[237] and the results of extensive studies at the University of Wisconsin on the effects of sodium fluoride on dairy cattle are available as a series of papers.[521,644-646,648,649,652] The results of one other large-scale study on the effects of sodium fluoride on cattle have been reported.[476] Experiments reported in the 1930's on the effects of fluoride fed as rock phosphate rather than as sodium fluoride, are also of value.[190,520,534]

Cattle have been the species most often affected by fluoride pollution; therefore, several reviews of fluoride toxicity in cattle are available. Roholm's monograph[549] reviews cattle effects up to 1937, and other reviews are available.[336,460,513] A critical review by Schmidt and Rand[571] and an extensive review by Cass[129] are perhaps the best sources of references to the literature that pertains directly to the effects of fluorides on livestock. Alther[24] has extensively reviewed the more recent European literature on fluorosis in livestock. The National Research Council publication, *The Fluorosis Problem in Livestock Production*,[519] is a very useful interpretive and authoritative report on the effect of fluorides on domestic animals.

The treatment of fluoride toxicity in most standard veterinary toxicology publications is rather superficial. A recent discussion of the effects of fluorides on cattle by Shupe and Alther[586] is more extensive but draws too heavily on the results of the Utah State experiments to be considered an interpretive review. A limited number of surveys of damage by fluorides in particular geographic areas from industrial or other sources have also been published.[17,76,116,451,684,729] Some are careful and extensive reports; others are very superficial.

Reviews directed specifically toward fluoride toxicity in livestock as an air-pollution problem have been prepared by Suttie.[638–640]

SOURCES OF FLUORIDE AND MODES OF EXPOSURE

Experimental Animals

The effects of fluorides on animals have been studied by the use of all conceivable routes of administration. Chronic effects have most often been studied by adding fluoride to the diet or the drinking water. It is helpful to keep in mind that, as a general rule, laboratory animals will consume about twice as many milliliters of water per day as grams of diet. In some studies, animals have been subjected to inhalation of various gaseous fluorides, and gastric intubation and intraperitoneal injections have been used, particularly in acute-toxicity studies.

Domestic Livestock

In the field, livestock are exposed to fluoride from the air, from ingested fresh forage or hay, from feed supplements, and from the water supply. There is also the possibility that high-fluoride soil, through rain splash or windblown dust, may contaminate forage.

It is usually assumed that the inhalation of fluoride from the atmosphere contributes a negligible amount to the total fluoride intake of animals in an area of industrial pollution. However, no available literature deals with the question of the hazard to animals of inhalation of fluoride from the ambient air. Rough calculations of the amount of air inhaled per day by a cow of average size indicate that the amount of fluoride that can be absorbed by this route is very small. Data on human inhalation of fluoride, in the form of hydrogen fluoride, reviewed by Largent,[350] also tends to support that contention. Inhalation of an atmosphere with a fluoride concentration of around 3 mg/m^3 for 6 hr/day resulted in fluoride absorption of 0.4–0.8 mg/kg (calculated on the basis of urinary excretion). Inasmuch as the concentrations of fluoride in the ambient air around industrial sources are in the micrograms-per-cubic-meter range (three orders of magnitude lower than that studied by Largent), it is doubtful that inhalation can contribute significantly to the total fluoride load.

The major source of fluoride in the ration of livestock in areas with a fluoride pollution problem is the ingestion of high-fluoride vegetation. The factors that affect the accumulation of fluoride in vegetation and the attempts to correlate ambient fluoride concentrations with accumulation in vegetation have been summarized in Chapter 7. The fluoride content of forage crops in areas subject to atmospheric fluoride pollution has been found by most investigators to be extremely variable. An example of this variability can be seen in the data presented in Table 8-1. It is apparent that in any given location, the fluoride content of the forage may vary by as much as tenfold from season to season. In general, the fluoride content is less during the early part of the summer, when the forage is growing rapidly, and rises during the fall. During periods of rapid growth, the accumulation of dry matter causes a continual dilution of fluoride. During the late fall or during a drought, dry-matter accumulation slows down and may even decrease. The reluctance of cattle to graze dry unpalatable forage also influences the fluoride content of their tissues from this source. Because forage that is cut and dried for hay represents the rapid vegetative growth of the early part of the summer, its fluoride content is more like that of the low-fluoride spring pasture than that of the fall and winter pasture.

TABLE 8-1 Monthly Variation in Pasture Fluoride

	Fluoride Content, ppm (Dry-Matter Basis)				
Month	Field I[a]	Field II[a]	Field III[b]	Field IV[c]	Field V[c]
January	125	116	–	110	89
February	–	–	–	84	47
March	154	118	–	55	70
April	53	80	41	63	48
May	118	58	23	101	74
June	25	33	8	12	14
July	84	134	19	19	12
August	84	63	16	16	11
September	84	72	34	14	7
October	102	72	30	20	17
November	220	174	42	16	69
December	292	150	–	117	67

[a] Data from Allcroft *et al.*[21]
[b] Data from Benedict.[44]
[c] Data from Huffstutler.[299]

Most reviews have indicated that natural forage fluoride contents are about 5–10 ppm (mg/kg, dry weight), but few extensive or systematic compilations of analyses are available. Suttie[641] has recently analyzed 107 samples of alfalfa hay from areas throughout the United States that were thought to be free of industrial fluoride pollution. The concentration of fluoride ranged from 0.8 to 36.5 ppm, with a mean of 3.6 ppm and a median of 2 ppm. There was no apparent relation between fluoride content of alfalfa and geographic location.

There is general agreement that there is little or no relation between the total fluoride content of soil and the fluoride content of plants grown in it. There is, however, some indication that acid soil promotes fluoride uptake and that liming of acid soil will reduce the fluoride content of crops grown in it.

Commercial feed supplements and mineral mixtures contain phosphates that may or may not be sufficiently high in fluoride to increase the fluoride intake appreciably. Documented cases of fluoride toxicity that have developed from the ingestion of meal or mineral supplements with an excessive fluoride content have been reported by Dale and Crampton.[166] Mortenson *et al.*[466] have published a method for estimating the fluoride intake of dairy cows in field conditions. The calculations provide a good example of how the relative contributions of various components of the ration to the total fluoride intake can be determined.

The fluoride contents of 168 samples of 16% dairy feed from seven different states have recently been analyzed by Suttie.[641] Although 90% of them contained less than 30 ppm of fluoride, some samples had over 200 ppm. It is clear that the contributions of fluoride from sources other than the ambient air must be considered in assessing field problems.

Although a large number of potential sources of fluoride emission have been listed, the sources that have been shown to be responsible for a toxicity problem in livestock appear to be limited to the aluminum, steel, phosphate, and brick industries.

Evidence of fluoride-induced changes in domestic animals cannot automatically be assumed to be due to industrial pollution. The problem of forage contamination by windblown dusts of natural origin and by soil splash as a result of heavy rainfall has been investigated in considerable detail by Merriman and Hobbs.[450] They have shown that crops grown on high-fluoride soils in agricultural, nonindustrial areas of Tennessee often had fluoride contents of over 25 ppm–in some cases as high as 300 ppm. Rand and Schmidt[532] have reported a case in which mixed forages contained 15-25 ppm of fluoride, although the irrigation water that was used contained 6.2 ppm; and Neeley and Harbaugh[473] have reported a case of fluorosis caused by the ingestion of high-fluoride waters.

ABSORPTION, DISTRIBUTION, AND EXCRETION

The general problem of the absorption, distribution, and excretion of fluoride by mammals is considered, with similar information from human studies, in Chapter 9. The extremely large body of information available in this field has been the subject of a recent extensive review by Smith.[606]

Ruminants are the animals most often associated with a practical fluoride-pollution problem, and there is little reason to believe that they metabolize fluoride differently from other species. Using radioactive fluoride, Perkinson *et al.*[517] and Bell *et al.*[61] have shown that fluoride is very rapidly absorbed by ruminants and that at least part of the absorption is through the rumen wall. Recent studies of Simon and Suttie[594] indicate clearly that a single oral dose of fluoride in the form of sodium fluoride is rapidly absorbed into the bloodstream of sheep and that the plasma fluoride concentration reaches a maximum in 1–3 hr (Figure 8-1).

There have been a number of attempts to decrease the absorption of fluoride in ruminants, and thereby lower its toxicity, by the addition of calcium or aluminum salts to the ration. The criterion of benefit has usually been decreased skeletal storage, and on this basis Hobbs *et al.*,[284] Greenwood *et al.*,[237] and Allcroft *et al.*[21] have observed a beneficial effect from aluminum sulfate, and Suttie *et al.*[645] have observed a beneficial effect from calcium carbonate. The degree of protection afforded by these additions has been variable, but there has usually been a 20–30% reduction in bone fluoride retention. There is no indication that farmers in an area of endemic fluorosis have attempted to alleviate the situation by the addition of these compounds.

There is no reason to believe that the factors that affect skeletal distribution and excretion of fluoride in ruminants are different from those in small experimental animals or man. However, very few studies have been set up to compare different species directly, with regard to any of these factors. In the absence of such data or of any real indication that they are needed, it can be assumed that the factors that influence fluoride distribution and excretion in man (discussed in Chapter 9) are also important in ruminants.

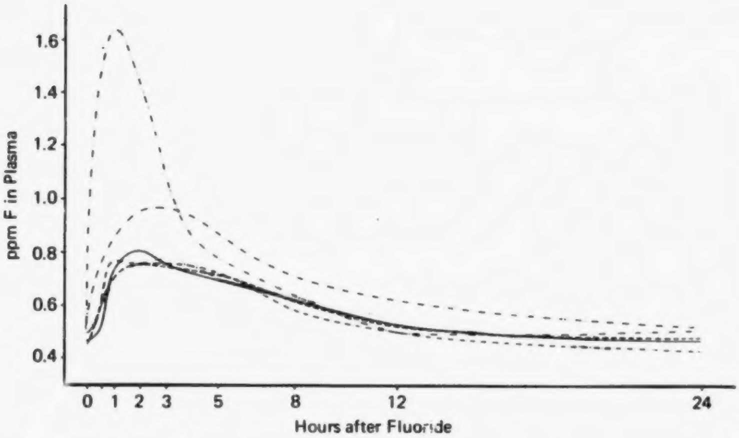

FIGURE 8-1 Diurnal variation in plasma fluoride. Each curve represents plasma fluoride response in single sheep given an oral dose of fluoride of 2 mg/kg of body weight at 0 time. (Reprinted with permission from Simon and Suttie.[594])

ACUTE EFFECTS

From the data compiled by Hodge and Smith,[287] it appears that the 24-hr intraperitoneal LD_{50} of a soluble fluoride, such as sodium fluoride or potassium fluoride, is about 10 mg/kg and that the oral LD_{50} for the same compounds is at least 5 times as great. The less soluble fluoride compounds are considerably less toxic, and the most toxic compounds studied have been beryllium oxyfluoride, indium borofluoride, and sodium borofluoride, which are about twice as toxic as sodium fluoride. At least in the case of rats given sodium fluoride, age appears to be an important factor, and younger rats are more resistant.

Acutely toxic doses cause gastroenteritis, muscular weakness, clonic convulsions followed by depression, pulmonary congestion (which may be accompanied by hemorrhage), and respiratory and cardiac failure.

The biochemical or physiologic basis of the lethality of fluoride is not clear. Enzymes involved in vital processes are inhibited, and severe hyperglycemia has been noted in some cases. Fluoride is a fairly effective inhibitor of cholinesterase, and this characteristic, with the decrease in plasma calcium concentration that has been observed, may be responsible for effects on the nervous system. Decreased calcium concentrations have also been postulated to affect blood clotting and membrane permeability. Cell damage and necrosis produce massive impairment in the function of vital organs, and, particularly when fluoride is given orally, there are severe local effects on the gastric and intestinal mucosa.

Information on cases of acute accidental and experimental poisoning of farm animals has been summarized and reviewed by Cass.[179] The symptoms are generally the same as observed in small experimental animals and man, but the data are insufficient to make any reasonable calculation of the minimal lethal single oral dose.

In a carefully controlled study with pregnant beef heifers, Hobbs *et al.*[284] attempted to feed 100–1,200 ppm of fluoride as sodium fluoride for 14 months. After 18 days of feeding, the animals receiving 600, 900, and 1,200 ppm were consuming so little of their feed that fluoride had to be administered in the form of a daily capsule. Food consumption remained low, and any animal that was too weak to rise for more than 2 consecutive days was slaughtered. The average periods for which the animals receiving 300, 600, and 1,200 ppm remained in the experiment were 130, 88, and 60 days. Animals receiving less than 300 ppm remained in the experiment for the entire 14 months. The symptoms seen in the animals were not those seen in acute single-dose experiments, but rather inanition and some of the skeletal changes observed in long-term studies.

CHRONIC EFFECTS ON DOMESTIC ANIMALS

Interest in the chronic effects of excessive amounts of fluoride in the ration of animals has centered on domestic animals. Although laboratory animals have been extensively used to study the absorption, distribution, and excretion of fluorides, interest in clinical symptoms has been directed mainly toward the animals in which a practical problem has been most evident—cattle. Because of this emphasis, the following description of the symptoms of fluoride toxicity deals mainly with cattle.

The chronic disease that arises from the ingestion of sublethal amounts of fluorides over an extended period and with which this review deals has been given different names by various investigators. "Fluorosis," "fluorine toxicity," "fluoride toxicity," "fluorine toxicosis," and "fluoride intoxication" have all been used, and there are no clear distinctions among them. Although "fluorosis" seems to have the most widespread use in reference to domestic animals, it, like the other designations, is rather ambiguous, in that it does not define a particular degree or severity of effect.

Signs of Excessive Fluoride Ingestion in Cattle

DENTAL LESIONS

Developing teeth are extremely sensitive to fluoride, and abnormalities in permanent dentition are the most obvious signs of the ingestion of increased amounts of fluorides in the ration. The histologic changes have been described and illustrated by Schour and Smith[575] and can be summarized as follows: The granoblasts (ameloblasts) are prematurely reduced in size and are often filled with hematoxylin-stainable globules. The epithelial papillae are abnormal, and the entire enamel epithelium frequently has a series of interruptions in its smooth surface, in the form of foldings or dips that extend into the enamel. This abnormal enamel matrix fails to achieve normal calcification, and the defect is later seen in the mature teeth.

The later effects of the histologic changes on the incisor enamel are generally described as chalkiness (dull chalk-like appearance), mottling (a pattern of chalk-like patches in the teeth), and hypoplasia (thin enamel). In addition, an affected tooth is subject to more rapid wear and in some cases to an erosion of the enamel away from the dentin.

The changes in the teeth can be given numeric scores based on severity, and a number of rather similar classification schemes have been described.[284,476,588,639] Color photographs showing examples of the various categories of tooth damage can be found in the NRC publication on fluorosis in livestock[518] and in the publication by Hobbs *et al.*[284] The somewhat simplified version of these schemes used by Suttie[639] is presented in Table 8-2.

Most investigators prefer viewing the defects in the enamel in transmitted light, which makes the early changes much more visible and almost makes it possible to ignore the degree of staining of the teeth. The amount of stain is an extremely variable expression of the defect in the enamel and can be very misleading. In all the classification systems proposed, a consistent factor has been that teeth with a score of 4 or 5 show definite enamel hypoplasia, whereas those with a score of 2 or 3 do not (Figure 8-2).

Cattle have three pairs of permanent mandibular incisors and one pair of permanent mandibular canine teeth. The canine teeth are commonly referred to as fourth incisors. In a careful study of young cattle of known ages, Brown *et al.*[110] have established the period of crown formation of the permanent teeth; their data are summarized in Table 8-3. The periods of tooth formation were determined by inspection of radiographs. Because the enamel matrix formation precedes the calcification process to some extent, the period during which

TABLE 8-2 Interpretation of Tooth Scores

Score	Appearance of Incisor
1	A normal tooth or one with a non-fluoride-caused lesion
2	Slight fluoride effects, usually horizontal mottling, seen best in transmitted light; surface of the tooth usually has a good luster
3	General mottling to the extent that the surface of the tooth has an overall chalky appearance
4	General mottling accompanied by hypoplasia of the enamel; may show increased wear
5	General mottling, hypoplasia, and wear to a greater extent than for 4; erosion of enamel from dentin

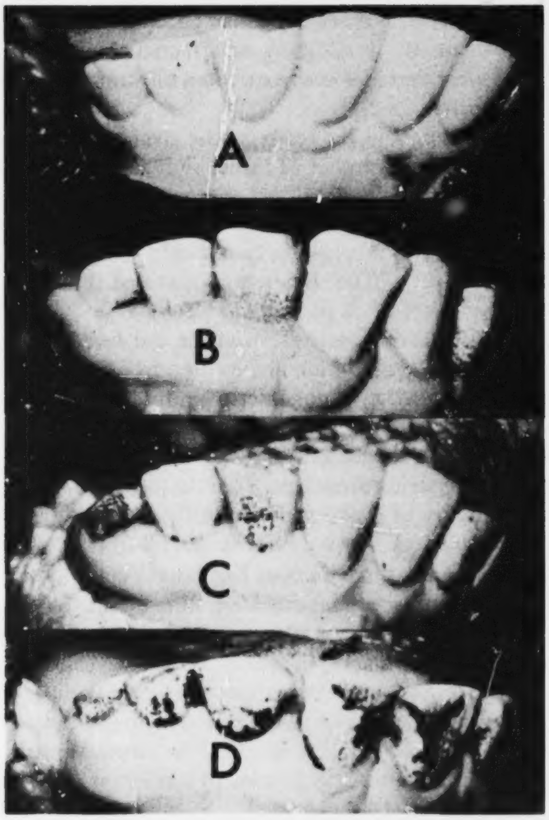

FIGURE 8-2 Examples of fluoride-induced changes in bovine incisor. Typical examples of types of lesions produced in permanent dentition by fluoride ingestion. Animals were first exposed to fluoride at 2–5 months of age. Fluorosis scores, reading from the first incisor to the canine tooth, are as follows: animal A, 1-1-1-1; animal B, 3-4-4-4; animal C, 2-4-2-5; animal D, 5-5-5-5. (Reprinted with permission from Carlson.[124])

the developing incisors are sensitive to fluoride should be considered to extend from the ages of a few months to about 30 months. Thus, animals not exposed to fluoride until they are 2½–3 years old will not show the typical fluoride-induced lesions. The molars are being formed

TABLE 8-3 Composite Chronology of Permanent Mandibular Tooth Development[a]

Tooth	Age at Beginning of Crown Formation, Months	Age at End of Crown Formation, Months	Age at Eruption, Months
1st incisor	6	12	23
2nd incisor	12	18	30
3rd incisor	20	24	36
4th incisor	28	33	42
1st premolar	13	23	24–30
2nd premolar	10	18	18–30
3rd premolar	11	27	30–36
1st molar	*(in utero)*	3	5–6
2nd molar	1	12	12–18
3rd molar	10	23	24–30

[a] Adapted from Brown *et al.*[110] Where no range is given, it is 1–2 months on either side of the average given.

during the same general period and, if subjected to increased exposure to fluoride, will also develop a defect that is later evident mainly as an increased rate of attrition. Garlick[224] has described the effects of fluoride on the cattle teeth and has discussed the uneven molar attrition line that results because of the more severe effects on one molar than on another (Figure 8-3). This effect is perhaps more important than the accelerated wear of the incisors. Inspection of the data in Table 8-3 reveals that the molars are all formed before the calcification of the fourth incisor. Therefore, a severe effect of fluoride on this incisor alone is not associated with excessive wear of the molars and is usually not considered to be as significant as effects on the other incisors.

OSSEOUS LESIONS

The ingestion of toxic amounts of fluorides for an extended period will result in the development of exostotic lesions and in some cases a general thickening of some bones. Grossly, the bones that exhibit this periosteal hyperostosis appear chalky white on the surface and are rough and porous, compared with normal hard smooth bones. In more advanced cases, mineralization of the tendons at the point of attachment to the leg bones has commonly been observed. Photo-

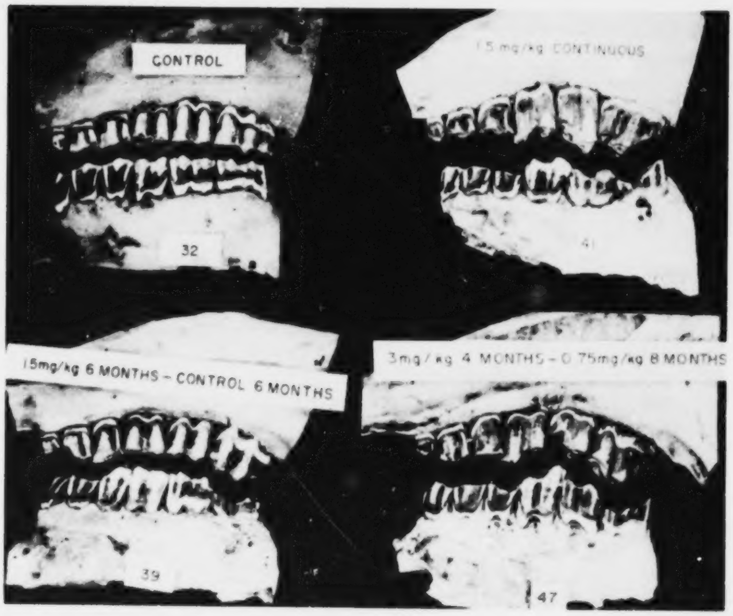
CONTROL
32
1.5 mg/kg CONTINUOUS
41
15mg/kg 6 MONTHS – CONTROL 6 MONTHS
39
3mg/kg 4 MONTHS – 0.75mg/kg 8 MONTHS
47

FIGURE 8-3 Effect of fluoride ingestion on wear of permanent bovine molars. Animals were fed indicated amount of fluoride from age of a few months to 6 years. Note even attrition line in control animal and uneven wear seen when fluoride was ingested during formative period. Detailed description of experimental procedure used can be found in Carlson.[124] (Reprinted with permission from Carlson.[124])

graphs of these lesions (Figure 8-4), which must be rather severe before they can be palpated with a measure of certainty, can be found in various publications.[116,588,652]

In addition to the exostosis commonly seen on the long bones, a thickening and outward twisting of the mandible leading to a flattened incisor arch[644] and excessive rib callus formation after rib fractures[116] have been described. The gross changes on the surface of the bones can also be observed by roentgenologic studies,[588] although it is doubtful whether this has much practical value as a diagnostic tool.

Accompanying the gross changes on the surface of the bone are extensive histologic changes. These appear to be very similar to those responsible for the production of abnormal enamel in developing

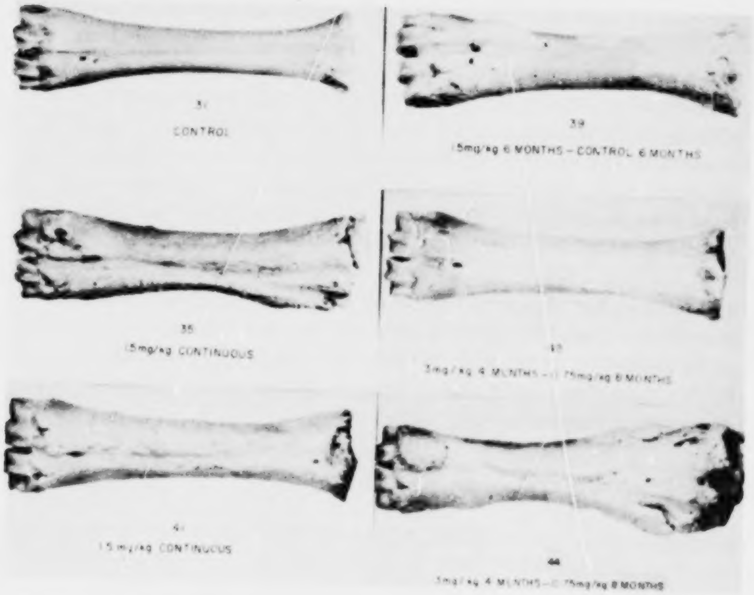
31
CONTROL
39
15mg/kg 6 MONTHS – CONTROL 6 MONTHS
35
15mg/kg CONTINUOUS
42
3mg/kg 4 MONTHS – 0.75mg/kg 8 MONTHS
41
1.5mg/kg CONTINUOUS
44
3mg/kg 4 MONTHS – 0.75mg/kg 8 MONTHS

FIGURE 8-4 Examples of fluoride-induced lesions on long bones. Examples shown are bovine metatarsal bones from cattle that were fed indicated amounts of fluoride from age of a few months to 6 years. Note generalized periosteal hyperostosis and prevalence of sharp exostotic lesions on some bones. Effects of fluoride ingestion on bones can be extremely variable; given level of intake does not have reproducible effect on bone. For details of the experimental conditions, see Carlson.[124] (Reprinted with permission from Carlson.[124])

teeth. The osteoblasts that produce the organic matrix are modified so that an abnormal matrix, which does not calcify properly, is produced . An excellent description of these fluoride-induced changes in bones of various species has been provided by Johnson.[323]

LAMENESS

The lameness associated with fluoride toxicity is usually described as a generalized stiffness and lameness and as being intermittent.[476, 588,645] It does not appear to be associated with the general periosteal hyperostosis, inasmuch as cattle with palpable exostotic lesions may appear to move normally. But it is likely that in advanced cases the calcifications of ligaments seen in some animals could influence movement. Of particular interest is the distinct type of lameness that is associated with a fracture of the pedal bone, or third phalanx, which, according to Burns and Allcroft,[116] occurs often in England. This fracture, which will usually heal in several weeks, results in a characteristic lameness and is thought to be prevalent in England because of the hardness of the ground in areas in which fluorosis is endemic. A number of pathologic changes of this particular bone have been described by Simon;[591] not all fractures of this bone should be ascribed to fluoride ingestion.

It must be stressed that animals become lame for many reasons and that, in the absence of other indications of excessive fluoride ingestion, a high incidence of lameness in a herd cannot be considered as an indication of fluoride toxicity. An excellent recent discussion by Amstutz[30] is a good source of descriptions of the various types of lameness that commonly afflict cattle.

CHEMICAL EVIDENCE

The most definitive evidence that animals are ingesting increased amounts of fluoride in their ration consists of chemical analysis of the forage consumed and an increase in the fluoride content of urine and body tissues.

The continual ingestion of fluoride results in a gradual accumulation of fluoride in the skeleton of animals as the fluoride ions replace hydroxyl groups and convert bone apatite to fluorapatite. The rate of accumulation of fluoride depends on the amount of fluoride in the ration. In four long-term experiments with dairy or beef cattle, in which up to 90 ppm of fluoride (dry weight) was added to the ration, the skeletal retention was found to be roughly proportional to the amount ingested (Figure 8-5). The rate of increase of skeletal uptake decreases with time, and this was readily apparent in dairy cattle experiments in which serial biopsies were taken.[588]

The amount of fluoride in different bones of the bovine skeleton varies appreciably, with cancellous bones—such as the pelvis, frontal, rib, and vertebrae—having higher fluoride contents than the more compact metacarpal and metatarsal bones.[588,649] There is also a large variation in the fluoride contents of different areas within a long bone, such as the metacarpal or metatarsal;[647] the dense cortical bone has a much lower fluoride content than the periosteal surface or the diaphyseal portion. It is therefore extremely important that analysis of such bones be accompanied by a description of the

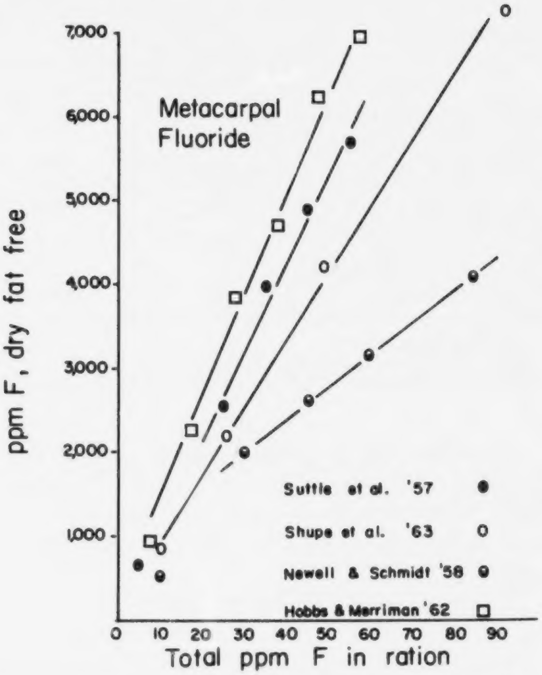

FIGURE 8-5 Relation between fluoride content of ration and fluoride content of metacarpal. Experiments conducted on a mg/kg basis have been converted to a ppm-in-total-ration basis. Period of exposure was not the same in all experiments. See Table 8-6 for details.

method of sampling and that the method of expressing the data (in terms of ash weight or dry fat-free weight) be clearly indicated.

In experimental conditions, in which there is a constant intake of fluoride, the amount of fluoride in the ration can be reasonably estimated if the skeletal fluoride content and the period of exposure are known. That cannot be assumed in field cases, as can be seen from the data of Mortenson *et al.*,[467] which clearly indicate widely differing degrees of dental damage in range cattle with similar intakes. Recent experiments (Table 8-4) on the controlled feeding of alternate high and low concentrations of fluoride do suggest, however, that skeletal analysis is a good indication of total fluoride intake, but not necessarily of the concentration of fluoride in past rations.[639]

TABLE 8-4 Effect of Variations in Pattern of Fluoride Intake on Skeletal Fluoride Retention[a]

Fluoride Intake, mg/kg of Body Wt[b]		Vertebral Fluoride Content, ppm Ash Wt[c]	
Actual	Yearly Average	1 Year	3 Years
1.5 for 12 months	1.50	5,420 ± 200	9,980 ± 1,850
1.5 for 6 months followed by 0 for 6 months	0.75	2,510 ± 230	5,130 ± 100
3.0 for 4 months followed by 0.75 for 8 months	1.50	5,385 ± 530	8,690 ± 680

[a] Data from Suttie.[639]
[b] Dairy heifers were started on the indicated intake at about 3 months of age.
[c] Fluoride analyses are means ± S.E. for four to seven animals per group.

Techniques for obtaining samples of skeletal tissue for analysis by vertebral biopsy have been published,[117,529] and Suttie[642] has established a correlation between the fluoride content of such bone and that of the metacarpal, which has often been analyzed in controlled experiments.

Urinary fluoride analyses are also used as an indication of fluoride exposure and are to some extent correlated with the dietary intake. The general relation between fluoride excretion and intake from a number of controlled experiments is indicated in Figure 8-6. Shupe *et al.*[587] have published a study relating the urinary fluoride content of cattle to fluoride intake and age. These correlations were established under conditions that included feeding of a constant amount of fluoride over an extended period, and they cannot be considered valid in field cases, in which intake may vary appreciably from day to day. Urinary fluoride concentrations will remain high for some time after removal of animals from a high-fluoride ration,[21,76,521] and they are therefore not the best indicator of current exposure.

As with other species, the ingestion of fluoride by cattle leads to a distribution of small amounts of fluoride in all tissues,[61] and slight increases in the fluoride content of various soft tissues have been seen in controlled experiments.[588,652]

Careful studies of plasma fluoride concentrations have been hampered in the past by the lack of a good analytic method. Simple methods based on the diffusion of fluoride[599] or on use of the specific ion electrode[212] are now available, and recent studies[124] have shown that the plasma fluoride concentration is directly correlated

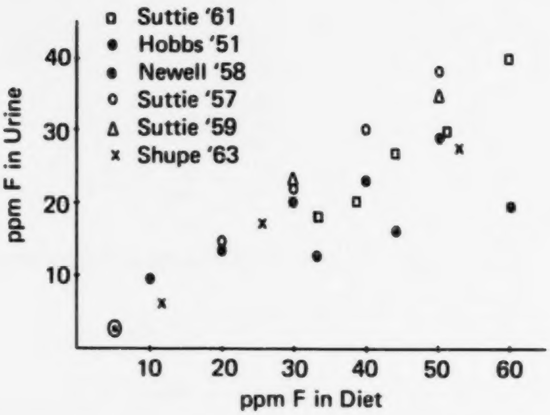

FIGURE 8-6 Relation between urinary fluoride concentration and fluoride content of ration. Data from all publications have been recalculated and corrected to a specific gravity of 1.040. Experiments conducted on a mg/kg basis have been converted to a ppm-in-total-ration basis. (Reprinted with permission from Suttie.[640])

with the degree of fluoride intake and subject to very rapid fluctuations as changes in intake occur. An illustration of the magnitude and rapidity of change of these values can be seen in Figure 8-7. Because of these fluctuations and difficulty in analysis, the values are of little practical use in diagnosis.

SYSTEMIC EFFECTS

Appetite impairment–eventually resulting in diminished milk yield, decreased weight gain, and cachexia– is an important factor in the development of the advanced stages of fluoride toxicity, but, because it is common to so many deficiency and toxic states, it is not in itself a good diagnostic criterion. Depending on the nature of the ration, the severe molar wear in advanced cases may also have some effect on food intake or digestibility. There is no evidence that fluoride ingestion can directly influence the physiologic process of milk secretion, and any effect on productive ability noted has been found to be secondary to other responses.

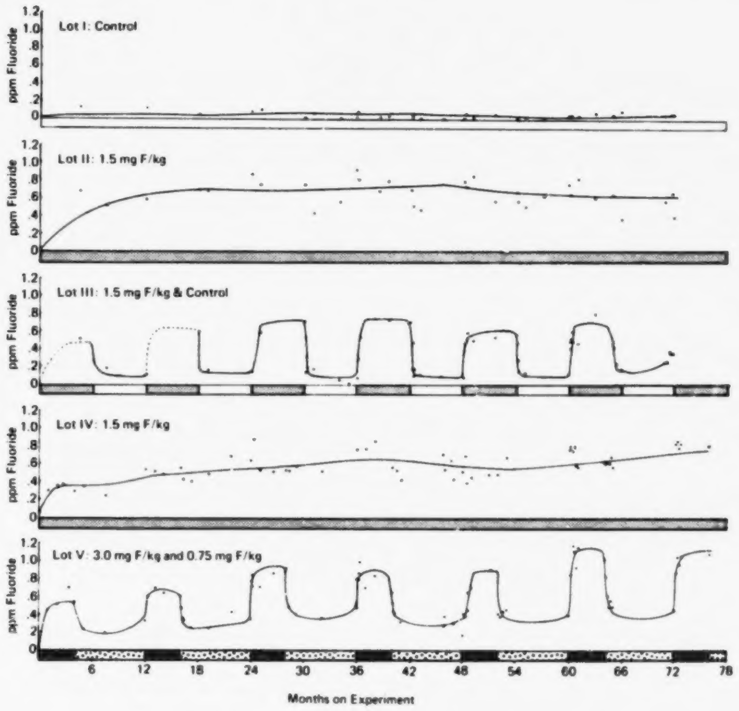

FIGURE 8-7 Relation between plasma fluoride concentration and concentration of fluoride in ration. Each point on graph represents average of number of animals in each experimental group. In groups in which fluoride concentration was changed on periodic basis, darkest crosshatching of bars represents period of highest fluoride exposure. See Carlson[124] for details of experimental conditions. (Reprinted with permission from Carlson.[124])

Stoddard *et al.*[633] found that 93 ppm of fluoride in the ration reduced feed consumption of dairy cattle after 2.5–3 years, but that 49 ppm resulted in no significant decrease during a 7.5-year experiment. In the same experiment,[632] it was found that after prolonged exposure, the lactating ability of some animals fed 93 or 49 ppm in the ration was adversely influenced. There was, however, no statistically significant effect of fluoride concentration on total production during the experiment. In a rather similar 5.5-year experiment, Suttie *et al.*[646] found no generalized effect on milk production when up to

50 ppm of fluoride was added to the ration, but they noted that some cows in their third or fourth lactation experienced a sharp temporary reduction in production that was concurrent with anorexia, weight loss, and lameness. The same group[649] found that mature cattle could tolerate 50 ppm for three lactations with no such response. Newell and Schmidt[476] noted no effect on production when up to 2.5 mg/kg (about 85 ppm in their experiment) was fed to heifers for 6 years beginning at the age of 2 years. In long-term studies with beef cattle, Hobbs and co-workers[283,284] found that 48 ppm was the lowest fluoride concentration at which a significantly lowered feed consumption was evident. All these studies used sodium fluoride as the fluoride source.

OTHER EFFECTS

Almost every ailment to which cattle are subject has been claimed by someone to be a symptom of fluoride toxicity. A field investigation by Udall and Keller[684] put a great deal of emphasis on poor reproduction, diarrhea, and overgrowth of the hoofs as good indications of the disease. The extensive controlled studies dealing with fluoride toxicity that have been reviewed here have failed to confirm these observations, as has a rather careful investigation of field problems.[116,636] Specifically, the claims of excessive hoof growth often raised in field cases have not been confirmed in controlled experimental studies; Simon and Leemann[592] studied the problem directly and found no influence of fluoride. Rensburg *et al.*[536] have indicated some effect on reproduction when rather high concentrations of fluoride were given to cattle in their drinking water, but many of the details of the experiment are not yet available.

Careful studies of the blood chemistry and liver and kidney function of dairy cattle by Hoogstratten *et al.*[292] and of beef cattle by Hobbs and Merriman[283] indicate that up to 100 ppm of fluoride can be fed in the diet with no changes in blood morphology, inorganic constituents of the blood, or kidney or liver function. The same groups[261,284] have demonstrated that as much as 100 ppm in the ration does not decrease dry-matter digestibility.

Many symptoms commonly ascribed to fluoride ingestion in the field are undoubtedly due to poor nutrition and management in a particular area being investigated; the significance of this problem has been specifically discussed by Crampton.[157]

Effects of Excessive Fluoride Ingestion on Other Species

The often-cited table from the NRC publication on fluorosis in livestock production (Table 8-5) indicates that cattle are the animals most sensitive to fluoride.[519] It has been shown[260,284] that feeder lambs and yearling ewes can consume up to 100 ppm of fluoride with no adverse effects on growth or lamb production. In a series of experiments, Peirce[514–516] has shown that sheep raised from birth to 7 years of age are adversely affected by 20 ppm of fluoride in the water supply, whereas ewes exposed at 2 years of age and yearlings continued in the experiments for 3½ years could tolerate this amount. Because of the pronounced fluctuations in water intake of the sheep in the experimental conditions, it is difficult to calculate how much fluoride was consumed on a dry-matter basis.

Only limited information regarding fluoride toxicity in horses is available. The available studies have recently been reviewed;[586,590] the conclusion was that horses can tolerate somewhat more flouride exposure than cattle, but probably somewhat less than sheep. The dental lesions and the type of periosteal hyperostosis seen are similar to the changes seen in cattle, and photographs of the changes appear in the two reviews cited.

Swine and poultry not only are more resistant to fluoride toxicity than most other animals, but do not normally ingest an appreciable amount of forage. Rather, they consume cereal grains, which are much lower in fluoride content. The effects of fluoride ingestion on

TABLE 8-5 Safe Levels of Fluoride in the Total Ration of Livestock[a]

	Fluoride Content, ppm	
Animals	As Sodium Fluoride or Other Soluble Fluoride	As Phosphatic Limestone or Rock Phosphates
Dairy cow	30–50	60–100
Beef cow	40–50	65–100
Sheep	70–100	100–200
Swine	70–100	100–200
Chicken	150–300	300–400
Turkey	300–400	–

[a] Adapted from Phillips *et al.*[519]

these animals, perhaps because of the lack of a practical problem, have not been subjected to the extensive study that has been given to effects in cattle. The best source of references to them is the review by Cass.[129]

The basis for the widely differing tolerance of different species is not known and has received little study. The resistance of the young chick to fluoride ingestion has been studied by Suttie *et al.*,[651] who have shown that the extreme tolerance is not due to an inability of the animal to absorb fluoride. Rather, the serum fluoride was found to be very high without apparent adverse effect on the animal.

Reports of the destruction of honey bees in the vicinity of fluoride-emitting industries have been reviewed by Lillie.[370] Although there is no doubt that bees have been killed, the available data are insufficient to determine what the toxic ambient fluoride concentrations or pollen fluoride concentrations were.

BIOLOGIC INDICATORS OF EXCESSIVE FLUORIDE INGESTION

The most definitive indication of excessive fluoride ingestion by animals is the steady increase in skeletal fluoride. The other chemical indices–urine and soft-tissue fluoride concentrations–are subject to short-term fluctuations and are not useful as a general indication of intake. Much the same can be said for the dental lesions, which represent the intake during the short time during which the teeth were forming, and the skeletal lesions, which appear to be subject to a great deal of individual variations.

The results of a series of cattle experiments at the University of Wisconsin (experiments A, B, and C in Table 8-6) suggested that concentration of fluoride in the long bones (metacarpal or metatarsal) in excess of 5,500 ppm (in terms of dry, fat-free weight) was indicative of and correlated with the appearance of systemic effects of fluoride toxicosis. Cows that had skeletal fluoride concentrations in this range also exhibited skeletal changes, excreted over 30 ppm of fluoride in the urine, were susceptible to lameness and appetite repression, and, if exposed to fluoride early enough, had badly mottled teeth of inferior wearing quality. It was suggested that the range of 4,500–5,500 ppm might be considered the marginal zone of toxicosis and that lower concentrations were not indicative of damage.

TABLE 8-6 Summary of Long-Term Experiments in Which Sodium Fluoride Was Fed to Cattle

	Experiment						
	A[a]	B[b]	C[c]	D[d]	E[e]	F[f]	G[f]
Initial age, years	2	4	1/3	¼	2	1	1½
Length of experiment, years	5½	3	4	7½	5½	10	8
Highest fluoride concentration fed, ppm[g]	54	54	90	93	85	58	108
Response: lowest level of fluoride, in ppm, in experiment to give response							
Decrease in growth rate	NE	NA	NE	NE	NE	NE	NE
Significant decrease in total milk production	NE	NE	NE	NE	NE	NA	NA
Interference with individual lactations	54	NE	NE	49	NE	NA	NA
Food intake repression	NM[h]	NM	NM[i]	93	NM	48	58
Tooth score of 4	34	NA	55	49	45	38	48
Tooth score of 2	24	NA	35	27	33	18	28
Moderate periosteal hyperostosis	44	54	55	49	60	48	58
Significant incidence of lameness	44	NE	55	49	85	NE	NE
5,000 ppm of fluoride in skeleton at 5 years[j]	54	NA	55	93	85	48	48
25 ppm of fluoride in urine[k]	34	34	55	93	85	38	38

[a] Data from Suttie *et al.*[645,646,652] and Suttie and Phillips.[648]
[b] Data from Suttie and Phillips.[649]
[c] Data from Suttie *et al.*[644]
[d] Data from Harris *et al.*,[261] Shupe *et al.*,[588] and Stoddard *et al.*[632,633]
[e] Data from Newell and Schmidt.[476]
[f] Data from Hobbs *et al.*,[284] and Hobbs and Merriman.[283]
[g] Total fluoride added (as sodium fluoride) plus amount in ration. In experiments C and E, intake was in terms of mg/kg, converted to ppm from data in original paper or from Stoddard *et al.*[633] and Suttie *et al.*[645]
[h] Temporary effect at 44 ppm.
[i] Temporary effect at 70 ppm.
[j] Based on metacarpal or metatarsal dry fat-free weight, in most cases extrapolated from other data in original references.
[k] Based on values taken after 2 years on experiment, expressed after correction to specific gravity of 1.04, except for experiment F, which is on an as-is basis.

NA = not applicable.
NE = no effect at highest level fed.
NM = not measured.

In the study of Newell and Schmidt,[476] 2-year-old heifers with continuous access to pasture were fed fluoride up to 2.5 mg/kg of body weight for an extended period and developed only mild physiologic symptoms of fluorosis. However, the skeletal fluoride concentration in animals that received the most fluoride averaged only slightly over 4,200 ppm, which indicates that they were only approaching the marginal area of toxicity. Similarly, in the extensive experiments of the Utah State group (experiment D in Table 8-6), it was only at the 93-ppm level—where excessive dental and bone lesions, serious lameness, and consistent systemic effects were noted—that the long-bone fluoride concentration exceeded 5,000 ppm. Bones of the animals that were fed 49 ppm were in the 4,000-ppm range. Caution must be taken in applying the results of these experimental studies to the conditions in the field, where the intake may vary greatly from one time to another.

No one criterion should be considered definitive of fluoride toxicity. Assuming that fluoride has been ingested from an early age, the first and most sensitive sign of toxicity will be the changes seen in the permanent incisors as they erupt. These changes do not appear to affect the functional value of the teeth until a damage score of 4 is reached. The signs may be followed by an increased incidence of lameness and the appearance of palpable bone lesions. During this time, chemical evidence of increased fluoride ingestion may be obtained from analysis of urine or skeletal biopsy; if the fluoride intake is high enough, there may be a temporary decline in milk production, which appears to be a secondary response to a decreased intake of food.

The extensive information available from experimental studies on cattle has been summarized in Table 8-6 in an attempt to give a clearer understanding of which chronic symptoms are associated with which fluoride concentrations ingested. No attempt has been made to include all the information available in the literature; rather, the data have been restricted to studies in which appropriate controls and methods were used and in which the numbers of animals involved were high enough to permit worthwhile comparisons. Particularly with regard to levels of ingestion that produce given tooth scores, there has often been appreciable individual variation within an experimental group, and the values listed should be considered as responses in typical animals ingesting the indicated amounts of fluoride.

ANIMAL RESPONSE IN RELATION TO AIR QUALITY

Criteria for the assessment of the potential effects of fluoride as a polluting agent in the atmosphere could presumably be based on any one of three characteristics: atmospheric fluoride concentration, forage fluoride concentration, and effects on the animals exposed.

The atmospheric fluoride concentration can be determined with available equipment and methods. However, the many factors affecting effluent dispersion would require continual monitoring over an extended period to get an accurate estimate of the average fluoride concentration in a given area. Even if ambient fluoride concentrations were known, such variables as the form of fluoride, growth rates of plants, and rainfall would make it difficult to extrapolate to forage concentrations or fluoride intake of cattle. What information is available on the relationship between ambient fluoride concentrations and forage fluoride concentrations has been discussed in Chapter 7.

The effects of fluorides on cattle can be assessed and evaluated and could presumably be used as a basis for criteria. However, because of the long latent period of many of the signs, such as dental markings, the damage might be done before the problem is recognized. The increases in fluoride contents of urine and plasma are indications of exposure but, as has been discussed, are too heavily subject to short-term variation to be useful as criteria. Fluoride analysis of a vertebral biopsy would stress the accumulative nature of the toxicity but does not seem feasible as a routine survey method.

Forage fluoride concentrations can be determined, although there are problems of collection and analysis (discussed in Chapter 4). Because of extensive information available on the effects of ingested fluoride and the correlations that have been made between ingestion level and various effects, the only rational basis for establishing criteria to protect grazing animals appears to be one based on the amount of fluoride accumulated in forage.

The carefully controlled experimental studies of the toxicity of fluoride in cattle have used sodium fluoride as a fluoride source. If the results of these extensive studies are to be used to establish criteria, the toxicity of fluoride in the form of vegetation contaminated with fluoride by industrial processes must be compared with the toxicity of fluoride as sodium fluoride. This is so important that assessments should be made not on the basis of studies in laboratory animals, but on the basis of cattle experiments.

Two well-controlled studies are available. Shupe *et al.*[589] have compared the relative effects of feeding fluoride as sodium fluoride, calcium fluoride, and hay contaminated with fluoride emitted by a steel plant on dairy heifers. They concluded that the contaminated hay was as toxic as sodium fluoride, but that the calcium fluoride was less toxic than either. In extensive beef cattle experiments, Hobbs and Merriman[283] found that fluoride in the form of rock phosphate was considerably less toxic than sodium fluoride and, on the basis of dental effects and skeletal fluoride retention, that fluoride in the form of contaminated hay from an aluminum smelter was somewhat less toxic than sodium fluoride. Because of the importance of these studies, the results have been summarized in Tables 8-7 and 8-8. An overall conclusion that can be drawn is that there is no indication that fluoride from an industrial source is any more toxic than sodium fluoride and some indication that it is slightly less toxic.

SUMMARY

Table 8-9 summarizes the relation between total fluoride content of forage ration and the development of various symptoms in livestock.[638] The statement "yes" or "no" should be interpreted as meaning that a given sign would or would not be expected to be reproducible at the indicated level of fluoride intake in the cattle. "No" does not mean that the sign could never be seen or has never been seen at that level, but rather that the incidence of its occurrence would not be expected to be significant. The table assumes a relatively constant intake of fluoride over a period of years by cattle first exposed as heifers to a fluoride salt with the same relative toxicity as sodium fluoride.

In discussing what effects on cattle should be protected against, there appear to be three general categories that can be defined—economic effects, severe dental fluorosis, and nondamaging evidence of fluoride ingestion.

Economic Effects. Carefully controlled experiments, extending over many years, have failed to demonstrate a statistically significant reduction in milk production when as much as 90 ppm of fluoride is included in the ration of dairy cattle. Various investigators have, however, indicated that, after 3–4 years of exposure at a level of about 50 ppm, the lactation of some animals within an experimental group did seem affected. The lack of a generalized response is consistent with the observation that a ration fluoride concentration in excess of 50 ppm is needed to influence food intake significantly. The lameness often seen at this concentration would not be expected to influence the animals' ability to graze or to consume dry forage.

TABLE 8-7 Comparison of Relative Toxicity of Fluoride in the Form of Contaminated Hay, Sodium Fluoride, and Calcium Fluoride[a]

Measurement	Type of Hay (Fluoride Concentration) Low Fluoride (10 ppm)	High Fluoride (62 ppm)	Low Fluoride + Calcium Fluoride (69 ppm)	Low Fluoride + Sodium Fluoride (68 ppm)
Average daily fluoride intake, mg/kg	0.20	1.14	1.32	1.29
Blood fluoride content, ppm	0.08	0.28	0.14	0.31
Urine fluoride content, ppm	4	15	11	20
Periosteal lesions, arbitrary score	0.10	1.90	0.42	1.92
Rib fluoride content, ppm, dry fat-free basis	530	3,860	1,430	3,731
Dental classification, fluorotic score				
1st incisor	0.75	2.00	1.25	1.75
2nd incisor	0.50	3.75	2.00	3.50
3rd incisor	0.50	3.00	1.00	3.00

[a] Data from Shupe *et al.*[589] Dairy heifers, 6–8 months old, were fed the different rations for 588 days. There were four animals per group. The measurements were taken near or at the time of termination of the experiment.

TABLE 8-8 Comparison of Relative Toxicity of Fluoride in the Form of Contaminated Hay and Sodium Fluoride[a]

	Type of Hay (Fluoride Concentration)	
Measurement	Contaminated Hay	Low Fluoride Hay + Sodium Fluoride
Average daily fluoride intake, mg/kg	0.87	0.86
Rib Fluoride content after 5 years, ppm ash	4,130	6,700
Urine fluoride content, ppm	18.1	19.6
Dental classification, fluorotic score		
1st incisor	2.0	2.7
2nd incisor	4.0	4.0
3rd incisor	3.7	5.0
4th incisor	4.0	5.0

[a] Data from Hobbs and Merriman.[283] Beef heifers, starting at 1 year of age had been fed the different rations for 5 years before these data were obtained. There were three animals per group; the data are for only two groups out of a large study. The contaminated hay ranged from 40 to 60 ppm of fluoride, and the total fluoride content of the low fluoride hay + sodium fluoride ration was 37 ppm.

TABLE 8-9 Relationship between Total Fluoride Content of Ration and Development of Various Signs in Livestock[a]

	Total Fluoride in Ration, ppm			
Symptom	20–30	30–40	40–50	>50
Discernible dental mottling[b]	yes	yes	yes	yes
Enamel hypoplasia (score 4)[b]	no	no	yes	yes
Moderate periosteal hyperostosis	no	no	yes	yes
Urine fluoride content equivalent to 25 ppm[c]	no	no	yes	yes
Significant incidence of lameness	no	no	no	yes
Decreased milk production	no	no	no	yes
Skeletal fluoride content equivalent to 5,000 ppm at 5 years[d]	no	no	no	yes

[a] Modified from Suttie[639]

[b] Only if fluoride is present during formative period of tooth.

[c] Based on values taken after 2–3 years of exposure; specific gravity, 1.04.

[d] Metacarpal or metatarsal bone, dry fat-free weight basis.

Severe Dental Fluorosis. Although there may be minor variations in the criteria by which different investigators have scored fluoride effects on teeth, in all scoring systems a tooth score of 4 represents the first category in which enamel hypoplasia is seen and in which an increased rate of incisor and molar attrition is expected. The concentration of fluoride needed to produce this effect has varied somewhat in different experiments, but, if the criterion is the production of only one pair of teeth with a score of 4 in a given animal, the concentration has tended to average 40–45 ppm. Teeth scored 2 or 3 have maintained a normal rate of wear for years after they erupted, whereas teeth scored 4 or 5 have worn abnormally fast.[283] At a constant level of fluoride intake, the canines (or fourth incisors) are usually the teeth most severely affected by fluoride. Because hypoplasia in these teeth is not associated with an increased rate of molar attrition, most investigators have tended to minimize its importance, and an animal should not be considered to have severe dental fluorosis if the canines are the only teeth affected.

Nondamaging Evidence of Fluoride Ingestion. At ingestion levels much below those required to produce a tooth score of 4, a trained observer can detect changes in dentition and there will be chemical evidence of an increase in the fluoride content of skeleton, urine, and soft tissues. Significant lameness or skeletal abnormalities will not be seen at levels below those which cause severe dental fluorosis, although histologic changes[323] might well be seen at very low levels of intake.

Suttie[639] has suggested that an air-quality standard should ensure that animals in an area suffer no economic damage and that there will not be a significant incidence of severe dental fluorisis in animals. There seems to be no justification for setting a standard that will prevent discernible deviations from normal that do not influence the general health, productive ability, or dental soundness of the animals.

The data summarized in Table 8-9 are based on total fluoride in the ration, and it should be remembered that, if a standard based on forage fluoride concentration is derived from these data, the total intake will be influenced by the amount of concentrate included in the ration. Some examples of the magnitude of that effect are given in Table 8-10, where the effects of forage with a fluoride content of 40 ppm and a concentrate with 10 ppm are considered. It should also be noted that most of the fluoride in the concentrate could be

TABLE 8-10 Examples of Total Ration Fluoride with Consumption of 40 ppm in Forage and 10 ppm in Concentrate

	Age, months	Weight, lb	Forage intake, lb	Concentrate intake, lb	Fluoride in total ration, ppm
Heifer	6-12	550	11	4	32
Heifer	12-18	800	16	3	35
Heifer	18-24	1,000	20	2	37
Cow, dry	24-30	1,200	30	2	38
Cow, 30# milk	24-30	1,200	30	8	34
Cow, 60# milk	24-30	1,200	26	20	28

present as rock phosphate, which would be less available. The examples chosen in the table are for growing dairy animals from 6 to 30 months old—the general period of tooth sensitivity. Beef heifers being raised to replace cows might not receive this amount of concentrate, nor would young beef cows have their fluoride intake diluted by concentrate feeding to this extent.

It is likely to be very difficult to set standards based on the use of forage fluoride as the measure of pollution in areas whose soil has a high fluoride content.

The report by Hobbs and Merriman[283] on the influence of high-fluoride soils or water on cattle points out that very high concentrations of fluoride can be found in forage growing in areas with no industrial pollution if the soil is of the fluoride-bearing "phosphatic basin" type. The accumulation is usually due to collection of dust or soil from rain splash on plants rather than to uptake from the soil. In such areas, it may be unreasonable to use forage fluoride as a basis for a standard.

Because most feeder cattle and lambs are in the feed lot for a relatively short period, there is insufficient time for skeletal lesions to develop and become a problem, even at relatively high levels of fluoride intake. These animals are often sold before the eruption of permanent teeth, so that this also poses less of a problem. The concern is thus only that the fluoride content of the ration does not influence food intake or digestibility. Shupe[585] has suggested that rations used for finishing beef heifers and steers might contain up to 100 ppm of fluoride, and those for fattening lambs, up to 160 ppm. Whether it would be possible to develop a manageable standard that would in some instances allow intakes that could not be tolerated over the lifetime of an animal remains to be considered.

TABLE 8-11 Effect of Short Periods of High Fluoride Exposure on Tooth Scores[a]

Cow	Peak Fluoride Intake	Peak Fluoride Concentration, ppm[b]	Months per Year at Peak	Yearly Fluoride Average, ppm[b]	Tooth Score, Incisor Number 1	2	3	4
A	Control	–	–	–	1	1	1	1
B	1.5 mg/kg	70	12	70	5	5	5	5
C	1.5 mg/kg	70	6[c]	35	2	4	2	5
D	2.5 mg/kg	115	3[d]	29	1	2	5	2

[a] Derived from Suttie.[639]
[b] Calculated on estimated dry-matter intake of 600-lb heifer.
[c] Re-exposed to fluoride at alternate 6-month intervals.
[d] Only one exposure to fluoride, 16–18 months of age.

Some thought will also have to be given to the observations that short periods of fluoride intake that are much higher than the average intake for a year may produce some severely damaged teeth in an animal. The effects of these alternating exposures have been considered by Carlson[124] and Suttie and Faltin.[643] The problem is illustrated by the data in Table 8-11, which contains examples from their studies. It can be seen that some severely affected incisors can be produced, even though the total intake over a year was in what would be considered a safe range. There is insufficient evidence to determine the length of time over which various fluoride concentrations can be tolerated without producing undesirable effects. On the basis of the available experimental information, Suttie[638] has made some suggestion of allowable ingestion periods and forage fluoride concentrations.

Although relatively little information has been published on animals other than cattle, the conclusion that can be drawn from the available information is that a fluoride-concentration limit that will protect cattle against damage will ensure adequate protection of other domestic livestock.

9

Effects of Fluoride on Human Health

Even a casual scanning of the fluoride literature soon convinces one that a multitude of effects have been produced, or are alleged to have been produced, by fluoride. It also becomes evident that a large proportion of the literature deals with experimental studies in animals, wherein the intent was to produce an effect for study, and that there is a paucity of good data relating to man. It is the purpose of this chapter to present information that will be helpful in assessing the effects of airborne fluoride on man. Accordingly, the greatest attention has been directed to reports that are believed to be appropriate to the purpose. That does not imply that other investigations and reports are not important, but rather that those selected are the ones thought to be helpful in dealing with questions that might arise in the field. Discussions of the efficacy of fluoride in reducing the incidence of dental caries and in treating some bone and hearing disorders are not included.

SOURCES OF FLUORIDE AND MODES OF EXPOSURE

Man obtains fluoride from a number of sources during his day-to-day activities, and in most instances these contribute to his intake every day. The three most important sources are water, food, and air; these vary in importance from one person to another and from time to time for a given person.

Generally, surface waters are low in fluoride and contain less than 1 ppm. However, in a number of localities around the world, the concentration may be as high as 10 ppm, or rarely 20 ppm or more. Summaries of reported fluoride concentrations in waters of Africa, the Americas, Asia, Australia, and Europe are presented in World Health Organization (WHO) Monograph 59.[353]

The subject of intake of fluoride from the diet and drinking water has also been reviewed in WHO Monograph 59[62] and by Hodge and Smith.[289] The observations of the latter authors are recapitulated and updated here.

Little fluoride is consumed in the diet, and the amount is nearly constant. The daily dietary (including drinking water) fluoride consumption in several countries has been estimated; it has been found to be at least 0.2 mg, and in only three has it been greater than 1 mg. The data are shown in Table 9-1. The fluoride concentrations in drinking water were also determined separately for most of the countries listed and found not to exceed 0.3 ppm. Some foods cooked in fluoridated water may undergo an increase in fluoride content.[410]

TABLE 9-1 Daily Dietary Fluoride[a]

Country	Fluoride Ingested in Food and Water, mg	Fluoride in Drinking Water, ppm
Canada	0.18 - 0.3	0.1
England	0.3 - 0.5	trace
Japan	0.47 - 2.66[b]	0.01 - 0.08[c]
Newfoundland	2.74[d]	trace
Norway	0.22 - 0.31	0.01 - 0.07
Soviet Union	0.6 - 1.2	0.2 - 0.3
Sweden	0.9	–
Switzerland	0.5[e]	–
United States	0.2 - 0.3[e]	–
	0.34 - 0.80	0.1

[a] Adapted from Hodge and Smith.[289]
[b] Including 0.07–0.86 mg from green tea.
[c] Milligrams of fluoride ingested.
[d] Including 1 mg from tea.
[e] Exclusive of that in drinking water.

The major food categories contain traces or more of fluoride, as shown in Table 9-2; the somewhat more recent data of Oelschläger *et al.*[494] are in general agreement. Meats generally are low in fluoride. Analyses of fish may show somewhat higher concentrations than meats, depending on whether the bones were included. For example, fresh fillets of cod, haddock, or herring contain 2–5 ppm, whereas whole eviscerated fish contain up to 57 ppm.[330] Sardines as consumed–i.e., including bones–may contain as much as 40 ppm. Fish-protein concentrate has been reported to contain 20–370 ppm;[330,750] but according to Zipkin *et al.*,[750] this fluoride is only one fourth to one half as available as sodium fluoride when the same concentration of fluoride from either source is incorporated into the diet of rats. Oelschläger *et al.*[494] point out that the commercial practice of raising trout in tanks may lead to somewhat higher fluoride concentrations in the fish because of the fluoride present in phosphates included in the ration. Fruits generally are low in fluoride (less than 1 ppm), and cereals and cereal products usually contain less than 3 ppm, as do vegetables and tubers.

Fluoride consumed in liquids comes chiefly from the natural fluoride content of drinking water or from fluoride added artifically as a caries-preventive measure. Beverages, with the exception of tea, do not contribute much fluoride (Table 9-3). Wines usually contain less than 1 ppm, although some have been shown to contain about 6 ppm. Tea leaves contain considerable amounts of fluoride, most of which is extractable by steeping or boiling: tea infusions generally

TABLE 9-2 Fluoride Concentrations in Fresh Foods[a]

Food, ppm	Data to 1959[b]	Recent Data
Meats	0.01 - 7.7	0.14 - 2
Fish	<0.10 - 24	≥1.0
Sardines	–	8 - 40
Shrimp	–	50[c]
Fish meal	–	186
Citrus fruits	0.04 - 0.36	0.07 - 0.17
Noncitrus fruits	0.02 - 1.32	0.03 - 0.84
Cereals and cereal products	<0.10 - 20	0.18 - 2.8
Vegetables and tubers	0.10 - 3.0	0.02 - 0.9

[a] Adapted from Hodge and Smith.[289]

[b] From Cholak.[139]

[c] Shrimp meat, 0.4 ppm; shrimp shell, 18–48 ppm.

TABLE 9-3 Fluoride Concentrations in Beverages[a]

Beverage	Fluoride Concentration, ppm
Wine	0.0 - 6.3
Beer	0.15 - 0.86
Tea infusion	0.1 - 2.0
Instant (soln.)	0.2
Coffee bean	0.2 - 1.6
Instant (powder)	1.7
Milk	0.04 - 0.55
Coca-Cola	0.07
Orange juice	0.0 - 0.05

[a] Adapted from Hodge and Smith.[289]

contain about 1 ppm. Fresh coffee beans contain up to 1.6 ppm; the decoction must be appreciably lower. Instant coffee contains about 0.02 ppm. Milk usually contains about 0.1 ppm. Coca-Cola and orange juice contain very little fluoride. The fluoride content of the water supply used in the preparation of any of these beverages will be reflected in the fluoride concentration of the final product.

It is evident that, unless a diet contains substantial amounts of particular seafoods or of tea, the fluoride content will always be low. When waterborne fluoride concentrations exceed 1 ppm, as they do in some areas, that source will become the principal contributor of ingested fluoride. The relative contributions of fluoride in water and in food to the total fluoride intake are seen in Table 9-4, which shows data obtained by Largent[350] for residents of communities whose water supplies contained up to 20 ppm of fluoride. Food intake averaged about 1.5 mg of fluoride or less, regardless of the fluoride content of the water. The amount of fluoride consumed in water, however, does depend on the concentration in the water, as well as on the amount of water consumed. (The data indicate that the adults observed each ingested approximately 1 liter of drinking water daily.)

The contribution to the total intake by fluoride that can be inhaled and retained can be highly variable. Thompson *et al.*[677] have reported that, of more than 11,300 24-hr samples of suspended particles collected over a 3-year period at urban and nonurban sites in the United States, over 90% contained less than 0.05 μg of water-soluble fluoride per cubic meter of air. If it is assumed that the average man doing moderately strenuous work inhales approximately

TABLE 9-4 Relation of Fluoride Ingested to That in Water and Food Consumed[a]

Fluoride in Water, ppm	Age, Years	Res., Years	Days Observed	Daily Fluoride Intake, mg		
				Fluid	Food	Total
<0.1	33	8	140	0.3	0.2	0.5
1[b]	–	–	–	–	–	–
2	35	10	96	2.4	1.2	3.6
5.5	55	29	60	3.8	1.3	5.1
6.1	57	34	133	6.7	1.0	7.7
8	57	19	140	11.3	2.5	13.8
20	30	8	45	20.8	1.5	11.3

[a] Adapted from Hodge and Smith.[289]
[b] Comparable data could not be found for persons consuming water containing 1 ppm of fluoride.

20 m^3 of air in a 24-hr period and that all the fluoride present is absorbed, the fluoride intake is readily calculated to be less than 1.0 μg, or less than 0.001 mg. Average concentrations of airborne gaseous fluoride of 1 and 3.5 ppb,* corresponding to the 1-month and 24-hr ambient fluoride standards, respectively, of New York State, would contribute about 0.002 mg or less than 0.06 mg of fluoride, assuming all of it to be absorbed. According to WHO Monograph 59,[409] fluoride inhaled by the average man in central London is about 0.003 mg in a normal day and may be 0.03 mg during a day of thick fog with exceptionally heavy pollution. Hodge and Smith[286] have estimated that a man working vigorously in an atmosphere containing the fluoride threshold limit value (TLV) of 2.5 mg/m^3, as established by the American Conference of Governmental Industrial Hygienists for industrial atmospheres (1969), would inhale 20-25 mg of fluoride during an 8-hr working day. If 25% of it were retained and absorbed, his daily fluoride intake would be 5–6 mg. Collings *et al.*[146] have reported that airborne fluoride-containing particulate rock phosphate and gaseous fluoride in equivalent fluoride concentrations are absorbed about equally well.

The possibility that fluoride may be ingested in anticariogenic agents—such as fluoride-containing tablets, lozenges, vitamin supplements, mineral supplements, dentifrices, chewing gums, and mouthwashes—should also be mentioned.

---

*Or 0.08 and 2.87 $\mu g/m^3$ of fluoride as hydrogen fluoride.

APPROXIMATE DAILY INTAKE

The preceding discussion permits two conclusions: that the daily intake of fluoride can be approximated and that such approximations cannot be as precise as one would like them to be.

- The diet exclusive of drinking water contributes at least 0.2 mg of fluoride to the total daily intake, and only rarely as much as 1 mg.
- The drinking water contributes an amount related to its fluoride concentration and to the volume of water consumed. For an adult in the temperate zone of the United States, the daily water consumption might be taken to be 1 liter, as a rule of thumb. On that basis, the fluoride contributed in milligrams per day by the drinking water is numerically equivalent to the fluoride concentration of the water in milligrams per liter.
- The ambient air contributes only a few hundredths of a milligram of fluoride, unless a person spends his working hours in an atmosphere containing fluoride in an amount approaching the threshold limit value. For example, a person employed in a workroom whose atmosphere contains 2.5 mg of fluoride per cubic meter (the current TLV) may retain as much as 5 mg of the inhaled fluoride, or even more.
- The use of fluoride-containing anticariogenic agents and other fluoride-containing drugs of supplements contributes variable amounts of fluoride to the total daily intake.

In summary, a rough calculation of the total daily fluoride intake of the adult is given by assuming 0.2 mg to come from food and adding an amount, in milligrams, equal to the fluoride concentration, in milligrams per liter, of the water supply consumed. For example, if a person resides in an area whose water supply contains 2 mg/liter, his total daily intake of fluoride can be estimated at 0.2 mg + 2 mg = 2.2 mg. The total intake would be somewhat greater if the person were using fluoride-containing drugs or dentifrices or if he were employed in an industry that released airborne fluoride.

ABSORPTION OF FLUORIDE

As yet, there is no convincing evidence that the absorption of ingested fluoride requires an active transport mechanism. The process is apparently passive, and it is rapid, as has been demonstrated repeatedly. Carlson *et al.*[123] orally administered 1 mg of fluoride labeled with fluorine-18 to human subjects, placed a portable scintillation counter over the abdominal region, and noted a rapid decline in radioactivity in the region of the stomach just a few minutes after ingestion of the dose. Other, indirect evidence of rapid absorption is afforded by the observation that fluoride appears promptly in the urine after it has been ingested. Data from a number of studies in man are plotted in Figure 9-1. Agreement among the several studies is reasonably good and indicates that approximately 20% of ingested soluble fluoride is absorbed and removed to the urine within 3 hr.

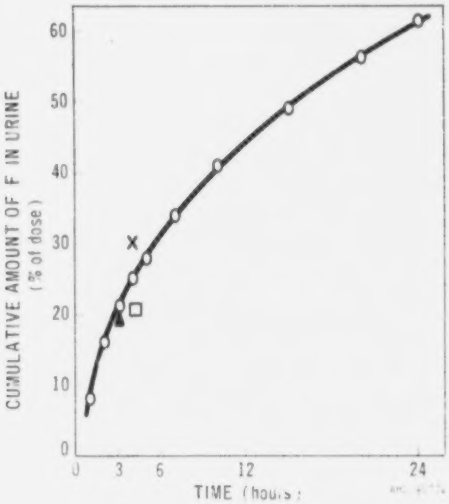

FIGURE 9-1 Cumulative excretion of fluoride in the urine of man after oral administration of sodium fluoride. ○, average from eight subjects drinking water containing 1 ppm of fluoride and given a single dose of 5 mg of fluoride.[743] Δ average from six subjects drinking water containing 0.06 ppm and given a single dose of 1.5 mg.[287 (p. 157)] X, average from two subjects drinking water containing 1 mg of fluoride labeled with ^{18}F.[123] ☐ average of seven trials with five subjects drinking water containing 1 mg labeled with ^{18}F.[195] (Reprinted from WHO.[731 (p. 151)])

Investigations into the site of fluoride absorption in the gastrointestinal tract so far have been limited to studies in rats, with the exception of the study of Carlson *et al.*[123] Although some of the experiments were performed under nonphysiologic conditions or used *in vitro* isolated segments of the tract, the evidence on the whole indicates that appreciable absorption of fluoride may occur across the stomach wall. Absorption is somewhat greater from the intestine and occurs throughout the first 15 cm of the rat intestine.

Absorption of fluoride entering the gastrointestinal tract is affected by a number of factors, such as the chemical nature of the ingested fluoride and the nature and extent of other components of the ingesta. Man usually absorbs at least 45% of the fluoride normally present in his diet[253] and may absorb nearly all of it[254,350] The effect on absorption of chemical differences in forms of fluoride is illustrated in Table 9-5. Bone meal is a poor source of fluoride. Sodium fluoride is extensively absorbed (97%), as are solutions of calcium fluoride and of cryolite. Calcium fluoride and cryolite in solid form are much less efficiently absorbed. Differences in solubility among solid forms are less important when small amounts are consumed.

Effects of various dietary components on the absorption of fluoride have been investigated in several animal studies. Ingestion of salts of calcium and of aluminum with fluoride has led to a reduction in fluoride absorption.[284] Magnesium apparently is effective at

TABLE 9-5 Fluoride Absorption in the Adult Human[a]

Form of Fluoride Added	Period, Weeks	Daily Fluoride Intake, mg	Fluoride Absorbed, %
Sodium fluoride in water	14	6.47	97
Sodium fluoride in water	2	12.40	97
Sodium fluoride in water	2	19.40	96
Calcium fluoride in water	4	6.25	96
Calcium fluoride, solid	3	6.43	62
Cryolite in water	5	25.40	93
Cryolite solid	4	6.41	62
Cryolite solid	3	6.61	77
Cryolite solid	3	12.40	67
Cryolite solid	6	18.40	70
Cryolite solid	3	36.40	65
Bone meal	5	6.31	37

[a] Adapted from WHO.[731]

TABLE 9-6 Effect of Calcium, Magnesium, and Aluminum on Fluoride Retention in the Rat[a]

Salt	Cation Concentration %	Total Fluoride Stored, mg[b]
$CaCl_2$	1.0	3.13
	0.1	5.92
	0.01	10.95
$MgCl_2$	1.0	5.88
	0.1	7.77
	0.01	11.43
$AlCl_3$	1.0	4.22
	0.1	8.49
	0.01	12.21
None	–	11.98

[a] Adapted from WHO.[731]

[b] 2 mg of fluoride (as sodium fluoride) administered daily by stomach tube. Total fluoride ingested: 24 mg.

concentrations of about 1,000 ppm or higher in the diet of rats, but not at 160 ppm or lower. Typical data are shown in Table 9-6. Manganese, molybdenum, and vanadium at dietary concentrations of 50 ppm did not affect fluoride retention by the rat.[731 (p. 87)] The significance of these observations with respect to human exposures to airborne pollutants containing these and other metals that may combine with fluoride is not known. High dietary fat has been shown to increase the retention of fluoride by rats.[118,455] In short-term experiments, Ericsson[194] noted an increased femoral uptake of fluorine-18 in rats that were also given flour or egg yolk. As mentioned earlier, the fluoride of fish-protein concentrate is about one fourth to one half as available as that of sodium fluoride.[750]

Collings *et al.*[146] have reported that in man fluoride is equally available in rock phosphate and in gaseous form, which seems surprising. Rye[564] has some evidence to the contrary.

DISTRIBUTION OF FLUORIDE

Blood

The quantitative measurement of fluoride in biologic materials is difficult at best, and its determination in plasma or blood seems to present the problem at its worst. Even a quick reading of the published data reviewed in WHO Monograph 59[731 (pp. 93-104)] and by Smith[606] reveals that values published by various authors over the years may differ by several orders of magnitude. Data accumulated from the literature by Smith are shown in Table 9-7. More recent work suggests that fluoride is present in human plasma in a nonbound and ionic form and in a bound form associated with albumin.[642,731 (p. 96)] The former fraction would be expected to be the portion participating in physiologic reactions. According to Armstrong and Singer,[731 (p. 96)] the ionic fluoride concentration is indicated by the observation that 15–20% of the total fluoride of normal human plasma is absorbed by calcium phosphate. In addition to the data listed here, Taves[664] has reported human plasma to contain an average of 0.013 ppm of fluoride. The consensus today of those currently engaged in research on plasma fluoride is that normal plasma fluoride concentrations in man are around a few hundredths of a microgram per milliliter. Singer and Armstrong[600] and Smith *et al.*[609] have presented evidence that some unidentified regulatory mechanisms operate to prevent widespread fluctuations in plasma fluoride concentrations in persons using water supplies containing approximately 0.01–2.5 mg of fluoride per liter. Data from animal studies, as reported by Carlson *et al.*,[123] Simon and Suttie,[593,594] Shearer and Suttie,[583] and Muhler *et al.*,[469] indicate that absorption from the diet produces slight and transitory changes.

Soft Tissues

In the last 30 years, fluoride concentrations in human organs have been reported from only five series of autopsies.[121,231,272,412,610] Data from the first three are summarized in Table 9-8. In each instance, there is no information with regard to the amounts of fluoride that the subjects ingested or were exposed to. Also, different analytic procedures were used. Consequently, it is not reasonable to make direct comparisons among the data. In Gettler and Ellerbrook's experience,[231] all organs analyzed contained less than 1 ppm of fluoride, whereas the data of Herman *et al.*[272] are consistently higher; indeed, Herman *et al.* state that all samples found to contain less than 5 ppm were reported as nil.

Table 9-9 lists the data of Call *et al.*[121] for fluoride in human tissues. The subjects were separated into two groups: "long-term," meaning persons over 15 years old who had resided for at least 10

TABLE 9-7 Normal Concentrations of Fluoride in Human Blood[a]

Fluoride Concentration in Blood, ppm	Comment	Reference
2.55 - 3.12	Adults	Agate *et al.*[17]
2.17 - 3.48	Children	
2.77	0.07 and 0.18 ppm in water supplies	Bowler *et al*[90]
0.04 - 0.36; avg., 0.18	Nonpregnant women; 0.55 ppm in drinking water	Gedalia *et al.*[238]
0.02 - 0.21; avg., 0.09	Pregnant women; 0.55 ppm in drinking water	
0.03 - 0.25; avg., 0.11	Cord blood of above pregnant women	
0.40 - 0.68	–	Gettler and Ellerbrook[231]
0.17 - 0.39	Pregnant women; 0.24 - 0.32 ppm in cord (fetal) blood	Held[264]
0.23 - 0.36	Normal hospital diet; 0.092 - 0.098 ppm in drinking water. Milk of these mothers contained 0.08 - 0.147 ppm	
0.19 - 0.24	Pregnant women	Held[265]
10 - 18	–	Matuura *et al.*[413]
0.14 (plasma)	0.15 ppm in drinking water supply	Singer and Armstrong[600]
0.15 (plasma)	1.1 ppm in drinking water supply	
0.19 (plasma)	1.1 ppm in drinking water supply	
0.16 (plasma)	2.5 ppm in drinking water supply	
0.26 (plasma)	5.4 ppm in drinking water supply	
0–0.09; avg., 0.014	0.06 ppm in water supply	Smith *et al.*[609]
0–0.09; avg., 0.04	1 ppm in water supply	
1.36	1 ppm in water supply; 1.02 ppm in urine	Yudkin *et al.*[737]
0.78	0.1 ppm in drinking water; 0.30 ppm in urine	
1.70	1 ppm in water supply; nephritics whose urine contained 0.64 ppm	
0.77	0.1 ppm in water supply; nephritics whose urine contained 0.32 ppm	

[a] Adapted from Smith.[606]

TABLE 9-8 Normal Concentrations of Fluoride in Human Organs[a]

Organ or Tissue	Fluoride Concentration, ppm in Fresh Tissue		
	Matuura *et al.*[413]	Gettler and Ellerbrook[231]	Herman *et al.*[b,272]
Bladder	–	0.40 - 0.68	15 - 185
Brain	1.8 - 4.3	–	–
Fat	3 - 4	–	<5 - 145
Gall bladder	3.9	–	–
Hair	14 - 30	–	13 - 171
Heart	1.3 - 6.6	0.44 - 0.60	–
Intestine	2 - 8	–	–
Kidney	2.1 - 94	0.56 - 0.78	<5 - 181
Liver	2.5 - 19	0.36 - 0.70	–
Lung	4.5 - 23	0.16 - 0.42	–
Marrow	44	–	–
Muscle	2 - 3.7	–	–
Nails	–	–	10 - 186
Nerve	16	–	–
Pancreas	2.8 - 38	–	–
Prostate gland	–	–	<5 - 86
Skin	5	–	10 - 290
Spleen	2.6 - 18	0.25 - 0.30	–
Stomach	2.9 - 6.7	–	–
Tendon	24	–	–

[a] Adapted from WHO.[731 (p. 86)]
[b] Concentration basis not stated; assumed to be fresh weight.

years (mean, 40.1 years) in an industrial area of Utah in which fluoride was released into the air; and "short-term," meaning persons under 15 years old who had lived for less than 10 years (mean, 2.32 years) in the same area. The dietary intake of fluoride presumably was approximately the same for all persons living in the area. Food crops grown in the area usually contained less than 0.5 ppm of fluoride, as did the water supplies (D. A. Greenwood, personal communication). The data of Table 9-9 show no major differences in fluoride concentration between the two groups; apparently, there was no significant storage over a longer period of exposure. The fluoride concentrations are somewhat higher than reported by Gettler and Ellerbrook[231] for "normal" subjects.

Table 9-10 shows fluoride concentrations found by Smith *et al.*[610] in tissues of persons whose water supplies contained 1 ppm of fluoride or less, or 1–4 ppm. The increased fluoride intake from water

TABLE 9-9 Fluoride Concentrations of Human Tissues[a]

Tissue	Fluoride Concentrations, ppm Fresh Weight			
	Normal[b]	Fluoride-Poisoning Fatalities[b]	After Exposure to High Atmospheric Fluoride Concentration[c]	
			Long-term	Short-term
Blood	0.27 (4)[d]	9.2 (5)[d]	–	–
Brain	0.53 (4)	2.5 (2)	1.8	1.5
Lung	0.27 (5)	14.0 (2)	3.9	3.5
Heart	0.45 (3)	10.6 (1)	2.5	1.9
Spleen	0.28 (2)	11.8 (1)	1.7	1.8
Liver	0.54 (3)	9.3 (5)	1.6	1.4
Kidney	0.68 (2)	9.0 (3)	2.9	2.9
Thyroid	–	–	5.2	4.0
Aorta	–	–	29.4	28.2
Pancreas	–	–	1.8	1.7

[a] Adapted from WHO.[731] (p. 101)
[b] Data from Gettler and Ellerbrook.[231]
[c] Data from Call *et al.*[121]
[d] Parentheses enclose numbers of subjects.

TABLE 9-10 Fluoride Concentrations of Human Soft Tissues[a]

Tissue	Fluoride Concentration (Dry-Weight Basis)	
	0–1.0 ppm in Water	1.0–4.0 ppm in Water
Heart	2.29 ± 0.796[b]	2.78 ± 1.364[b]
Liver	2.34 ± 0.972	2.27 ± 1.109
Lung	5.12 ± 3.104	6.18 ± 3.106
Kidney	3.26 ± 0.968	8.49 ± 6.63
Spleen	4.91 ± 4.143	3.53 ± 1.324
Aorta	41.0 ± 50.27	25.1 ± 19.54

[a] Adapted from WHO.[731] (p. 100)
[b] SD.

did not result in substantial increases in tissue fluoride concentrations. Both Call *et al.*[121] and Smith *et al.*[610] found fluoride concentrations to be greater in the aorta than in any other tissue examined; Call *et al.* noted that this increase paralleled the degree of aortic arteriosclerosis and calcification.

The distribution of fluoride within soft tissue is not fully known. However, both Partridge[511] and Armstrong and Singer[47] have indicated that at least a portion of the fluoride is to be found in the intracellular water.

Bone

The two mechanisms whereby the body disposes of absorbed fluoride are deposition in the calcified structures and excretion through the kidneys into urine. The importance of these processes is evident when it is considered that 99% of the fluoride retained in the body is localized in the skeleton and that ingested fluoride appears promptly in the urine, as much as 20% of it being found there in the first 3–4 hr after ingestion. Nearly all that absorbed is excreted via this route within 24 hr.

A number of investigations with fluorine-18 have demonstrated the rapidity of skeletal uptake. The data of Wallace[701] are appropriate for demonstrating the course of this uptake, and these data, obtained in rats after ingestion or intraperitoneal injection, are shown in Figure 9-2. Other data from Wallace's experiment indicate that the fraction of the dose appearing in the skeleton is 1.5–4.5 times greater than that appearing in the urine. Skeletal deposition, then, is the more rapid process, and its rapidity thus regulates the size of the fractions deposited and excreted.

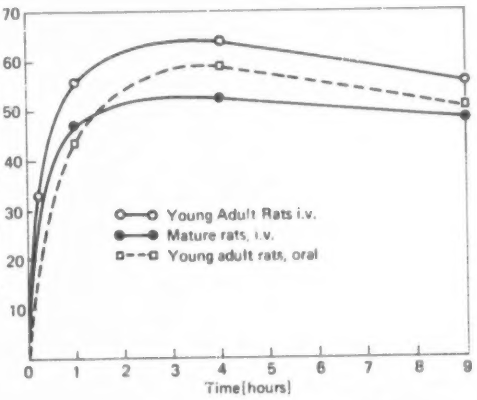

FIGURE 9-2 Uptake of ^{18}F by the rat skeleton. Data of Wallace.[701] (Reprinted with permission from Smith.[606])

A number of factors affect bone fluoride concentration. Consequently, the phrase "normal fluoride content" is without meaning unless surrounded by a rather awkward complex of qualifying phrases related to age, sex, present and previous fluoride intake, specific bone, part of bone, and perhaps other factors.

The concentration of fluoride increases with time, as several surveys have shown. For example, Smith *et al.*[608] analyzed the rib cortex of samples taken at autopsy of persons who, so far as could be ascertained, were lifetime residents of an area whose water supply contained approximately 0.06 ppm of fluoride. Their results are shown in Figure 9-3. The data of Jackson and Weidmann,[311] for persons using water supplies containing less than 0.5, 0.8, and 1.9 ppm of fluoride, are shown in Figure 9-4. Figure 9-3 suggests that fluoride in bone increases throughout life, whereas Figure 9-4 suggests a plateau level of fluoride concentration at about the age of 55. This seeming contradiction is discussed later.

Older animals store less of the daily intake than do younger ones, as is indicated by considerable experimental data. Several appropriate studies have been summarized in WHO Monograph 59[731 (p.110)] (see Table 9-11).

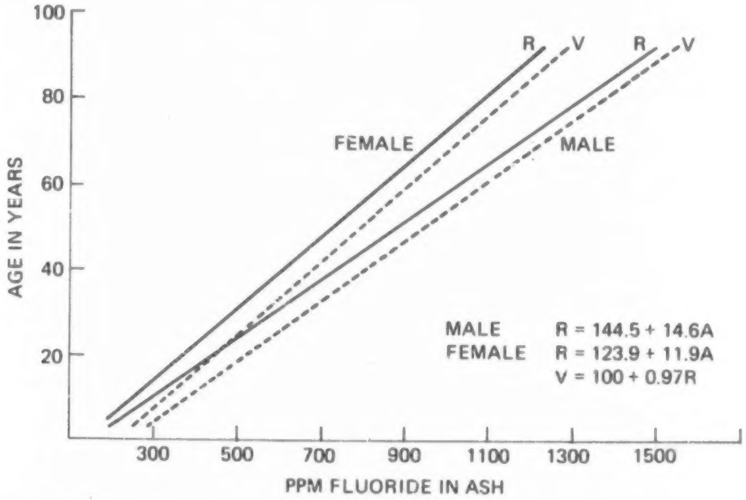

FIGURE 9-3 Fluoride concentrations in human bone ash of lifetime residents of a low-fluoride area. Data of Smith *et al.*[608] (Reprinted with permission from Hodge and Smith.[287])

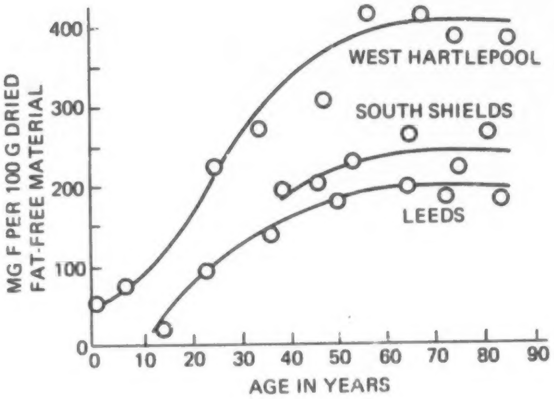

FIGURE 9-4 Relations among fluoride concentrations in bone, age, and fluoride in water supply. Data of Jackson and Weidmann.[311] Fluoride in drinking water: West Hartlepool, 1.9 ppm; South Shields, 0.8 ppm; Leeds, less than 0.5 ppm. (Reprinted with permission from Hodge and Smith.[287])

At all ages, some areas of bone are more active than others, and within an individual bone, the distribution of fluoride corresponds rather well with this pattern. Cancellous bone contains more fluoride than does compact bone, and the biologically active surfaces of bone take up fluoride more readily than does the interior. Weidmann and Weatherell have discussed this in WHO Monograph 59[731 (pp. 110-111)] and summarized pertinent data. Their summary table is reproduced here as Table 9-12. The effects of blood flow and vascularity on local deposition of fluoride have been reviewed in WHO Monograph 59,[731 (pp. 114-115)], as has the effect of crystal size and surface. In biologically active regions, there is an extensive and accessible surface area of many small hydrated crystallites available to the ions of tissue fluids. These differences in crystal growth, size, vascularity, and blood flow account for most of the observed variations of fluoride distribution.

As the fluoride concentration in the ration increases, the concentration of fluoride in the bone also increases. The effects of various components of the diet on availability of the fluoride have already been commented on. Inasmuch as the fluoride intake in the diet is reasonably stable and is often much less than that contributed by

TABLE 9-11 Retention of Fluoride by Bones of Growing and Mature Animals[a]

Reference	Species	Bone	Age	Dose	Duration, Days	Fluoride, ppm in Ash
Zipkin and McClure[746]	Rat	Femur	30 days	10 ppm in	136	1,240
			90 days	water until	139	1,140
			150 days	a total of	132	860
			210 days	20 mg	116	720
			330 days	ingested	107	470
Suttie and Phillips[650]	Rat	Femur	Weanlings	500 ppm in	3	2,333
			Young	food *ad lib.*		1,038
			Mature			771
Weidmann and Weatherell[707]	Rabbit	Long bones (diaphysis)	1–2 months	500 ppm in	22	6,180
			3–4 months	water *ad lib.*		4,100
			Mature			1,555

[a] Adapted from WHO.[731] (p. 110)

TABLE 9-12 Retention of Fluoride by Different Regions and Types of Bones[a]

Reference	Species	Age	Bone Type and Region	Dose	Duration	Fluoride, ppm in Ash
Gardner *et al.*[233]	Dog	Adult	Diaphysis, compacta	13 ppm in diet *ad lib.*	Life-long	2,285
			Epiphysis, cancellum			3,132
			Diaphysis, subperiosteal			3,257
			Diaphysis, interior			2,160
		Young	Diaphysis, compacta			–
			Epiphysis, cancellum			415
			Diaphysis, subperiosteal			345
			Diaphysis, interior			315
Weidmann and Weatherell[707]	Rabbit	Adult	Diaphysis, compacta	500 ppm in water *ad lib.*	22 days	1,585
			Metaphysis, cancellum			9,220
			Diaphysis, subperiosteal			6,640
			Diaphysis, average			1,585
		3–4 months	Diaphysis, compacta			4,070
			Metaphysis, cancellum			9,070
			Diaphysis, subperiosteal			5,680
			Diaphysis, average			4,070
		1–2 months	Diaphysis, compacta			5,860
			Metaphysis, cancellum			9,470
			Diaphysis, subperiosteal			7,860
			Metaphysis, average			5,860
Weatherell (unpublished data)	Man	33 years	Diaphysis, compacta	<0.1 ppm in water *ad lib.*	Life-long	703
			Metaphysis, cancellum			885
			Diaphysis, subperiosteal			1,250
			Diaphysis, interior			679
			Rib, compacta			1,685
			Rib, cancellum			3,500

[a] Adapted from WHO.[731] (p. 111)

the water supply, it is apparent that fluoride in the skeleton usually is more closely related to the latter source. This relationship is shown in Figure 9-5, based on data from the work of Zipkin *et al.*[748] and McClure *et al.*[423] The subjects represented here ranged in age from 26 to 90 years and used their water supplies for 10–87 years. As would be expected, skeletal concentration increases as exposure continues, although the rate of accretion may decline; these features of the process have already been illustrated in Figures 9-3 and 9-4.

A number of studies in animals have indicated that if a person is treated for a short time with doses larger than his normal intake, 50% or more of the absorbed fluoride is deposited in the skeleton.

If "steady state" is defined as the condition in which the total amount of fluoride in the bone does not change as additional fluoride is ingested, so that daily intake is exactly balanced by daily loss, then we have little evidence to demonstrate such a state.

The data of Smith *et al.*[608] shown in Figure 9-3 indicate that, at the low concentration of fluoride ingested, a plateau was not reached. Weatherell[704] has also presented data on fluoride in the compact bone of femora of persons residing in an area containing 0.1 ppm of fluoride in the drinking water. His data (Figure 9-6)

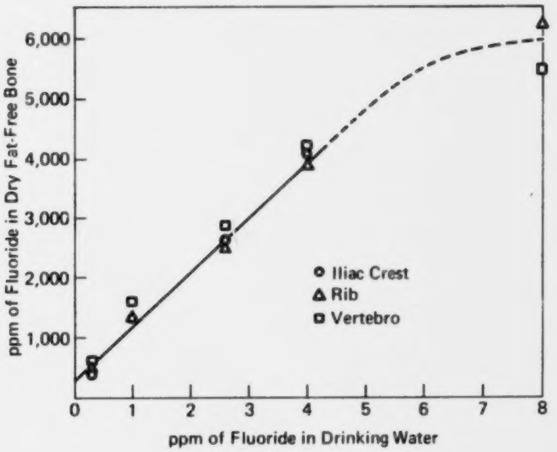

FIGURE 9-5 Relation between fluoride in drinking water and fluoride in human bone. Data of Zipkin *et al.*[748] and McClure *et al.*[423] (Reprinted with permission from Smith.[606])

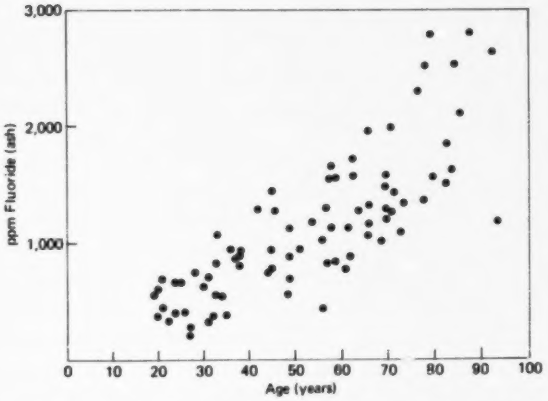

FIGURE 9-6 Fluoride concentration in cortical bone from midshaft diaphysis of human femora of different ages from areas supplied with drinking water containing less than 0.5 ppm. Data of Weatherell and Weidmann (unpublished). (Reprinted with permission from Weatherell.[704])

also do not indicate that the rate of fluoride accumulation declines with age. Jackson and Weidmann's data[311] (Figure 9-4) suggest that a plateau had been reached in their subjects, and Blayney *et al.*,[77] in their study of residents of Evanston. Illinois, ranging in age up to 80 years, suggest a tendency for fluoride accumulation in the iliac crest to level off or at least to proceed at a lower rate. The data of Zipkin *et al.*[748] do not suggest a lower rate of accretion. Evidently, more work is required before any categoric statement can be made about a plateau of fluoride accumulation in man.

Evidence from several animal studies has indicated a tendency for fluoride concentrations to reach a constant level. Representative data from a study of Zipkin and McClure[746] are shown in Figure 9-7. For the first 200 days, the fluoride content of the femora increased rapidly. Between 200 and 300 days, the fluoride accumulated more slowly; there was no further increase. It is difficult to say to what extent age of bone and prolonged or increased fluoridation contribute to the development of the equilibrium state; the decreased growth rate of bones with increasing age would account for a portion of the reduced rate of incorporation. Suttie and Phillips[650] also noted that, after an initial period on 500 ppm of fluoride in the diet, rats' bones accumulated less fluoride, irrespective of age.

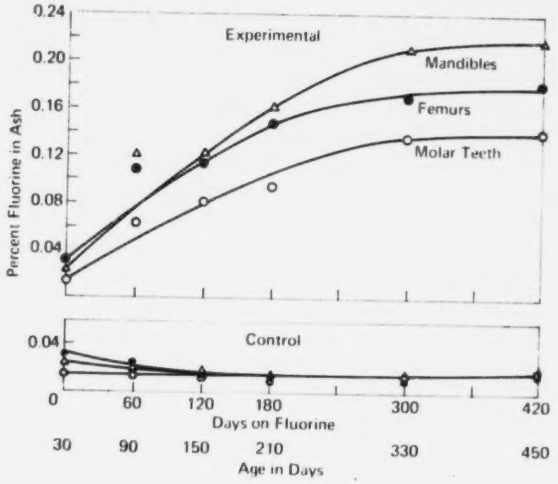

FIGURE 9-7 Fluoride concentration in bones and teeth of growing rats receiving 10 ppm of fluoride as sodium fluoride, per day, for 420 days, starting at 30 days of age. (Reprinted with permission from Weatherell.[704])

Fluoride probably increases more rapidly in bone not previously exposed to it. The absorption of fluoride might then saturate the available sites and thereby diminish uptake. Such saturation probably does not develop at very low levels of fluoridation, even after lifelong exposure. The normal physiologic processes of bone remodeling and renewal are probably sufficient to permit a steady incorporation of small amounts of fluoride.[704]

Figure 9-3 indicates that, in the human ingesting low concentrations of fluoride, bones of a female will contain more fluoride than those of a male of the same age.

Teeth

The subject of the incorporation of fluoride into teeth has been reviewed by Weidmann and Weatherell in WHO Monograph 59[731 (pp. 104-128)] and by Weatherell.[704] The salient points are recapitulated here.

The factors controlling incorporation of fluoride into dental structure are essentially the same as those pertaining to bone. Uptake is more rapid during growth and development. Once formed, however, the dental tissues, unlike bone, do not undergo remodeling, and there is no cellular activity in the enamel. Enamel has a low average fluoride concentration. Relatively low concentrations are achieved during the brief period of formation. When a minimal degree of calcification has been achieved, the increasing resistance to ionic penetration soon leads to a concentration gradient from the tooth surface to the interior. This preferential accumulation on the surface occurs before eruption, probably after the enamel has been formed but before calcification is complete. In the fully calcified permanent tooth, the rate of uptake is low. The change in fluoride concentration with depth in the tooth has been shown in several investigations in which successive layers of enamel have been removed and analyzed for fluoride. Typical of such data are those of Weatherell and Hargreaves,[705] reproduced in Figure 9-8.

The average concentration of fluoride in dentin is 2–3 times that in enamel and is affected by growth and mineralization. As is true

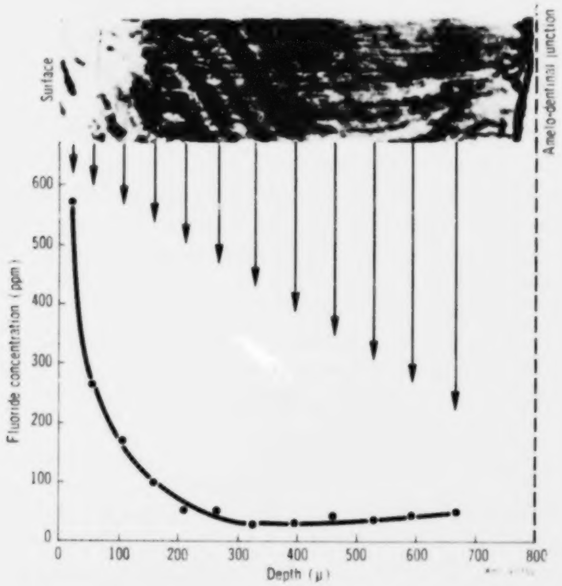

FIGURE 9-8 Pattern of fluoride distribution in human enamel in the labial aspects of permanent upper incisor. (Reprinted from WHO.[791 (p. 113)])

for bone and for enamel, dentinal fluoride is greater in the surface regions than in the interior. Concentrations are highest closest to the pulp chamber.

Cementum has been much less frequently studied. It is more closely related to bone than to enamel or dentin, and its fluoride content has been reported to be greater than that of bone.[595]

Fluoride levels of both dentin and enamel are related to the concentration of fluoride in the drinking water, as the data of Table 9-13 show.

As with bone, fluoride in the dentin and enamel increases with age, but the rate of accumulation gradually declines. The situation is shown graphically for dentin and for enamel in Figures 9-9 and 9-10, respectively, taken from WHO Monograph 59.[731 (pp. 126-127)]

Placenta and Fetus

The distribution of fluoride in the placenta and fetus has been reviewed by Gedalia in WHO Monograph 59.[731 (pp. 128-134)] The placenta allows fluoride to reach the fetal circulation to some extent, after which factors governing skeletal deposition during extrauterine life also affect skeletal deposition in the fetus.

Extraskeletal Sites of Calcification

It has already been mentioned that, of all organs and tissues analyzed for fluoride, only the aorta has shown unusually high concentrations of fluoride. Smith *et al.*[610] suggested that this could be associated with the increased calcification frequently seen in this tissue with increasing age, although calcium determinations were not made. Call *et al.*[121] did measure calcium concentrations in their series and showed that the fluoride content in the aorta did indeed parallel the changing calcium content.

Bernstein *et al.*,[70] during the course of a survey of the incidence of osteoporosis in residents of high- and low-fluoride areas (4–5.8 ppm and 0.15–0.3 ppm, respectively), found the incidence of calcified aortas to be significantly reduced in males in the high-fluoride area. A similar trend was also evident in the women of both areas. Korns[343] examined the x-ray films of residents of Newburgh (1 ppm of fluoride in the water) and of Kingston (0.05 ppm) and noted increased aortic calcification with age in both groups, but without significant differences in incidence between the groups.

TABLE 9-13 Fluoride Concentrations in Dentin and Enamel at Different Levels of Fluoride Ingestion[a]

					Fluoride Concentration, ppm in Ash			
Reference	Species	Age	Fluoride Dose, ppm in Water	Duration	Enamel Surface	Interior	Whole	Dentin (Whole)
McClure and Likins[422]	Man	Adult	0.1	Life-long	–	–	86	332
			7.6	Life-long	–	–	658	1,968
Jackson and Weidmann[312]	Man	20–49 years	<0.5	Life-long	–	–	108	508
		20–35 years	1.2	Life-long	–	–	180	922
		20–35 years	1.9	Life-long	–	–	320	1,290
Jenkins and Speirs[322]	Man	Adult	<0.25	Life-long	590	80	–	–
			1.4	Life-long	960	110	–	–
			2.0	Life-long	1,310	270	–	–
Brudevold *et al.*[113]	Man	20–29 years	0.1	Life-long	571	48	–	–
			1.0	Life-long	889	129	–	–
			3.0	Life-long	1,930	152	–	–
			5.0	Life-long	3,370	570	–	–

[a] Adapted from WHO.[731 (p. 117)]

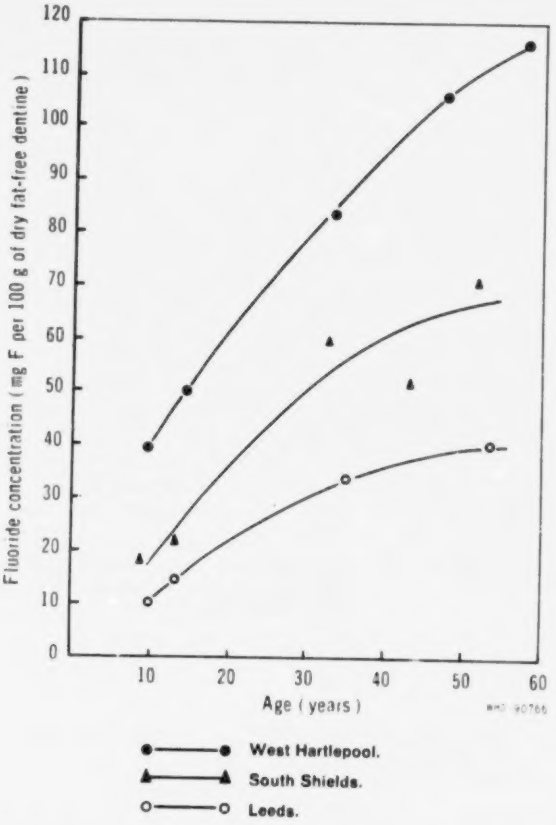

FIGURE 9-9 Relation between age and dentinal fluoride in human premolars. (Reprinted from WHO.[731] (p. 126))

The autoradiographic studies of Ericsson and Ullberg[197] have shown that fluoride is localized in areas of the placenta of mice coinciding exactly with those of calcium deposits.

Several investigations have shown fluoride to be present in renal calculi; concentrations in stones from persons whose water supplies contained 0.6 ppm of fluoride or less were not significantly different from those in stones from persons who used water containing 2.6 ppm. Some biliary tract calculi from the latter persons contained fluoride, and others did not.[743]

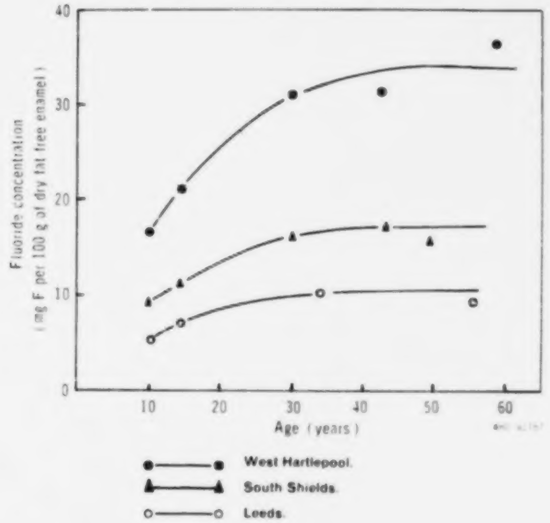

FIGURE 9-10 Relationship between age and enamel fluoride in human premolars. (Reprinted from WHO.[731 (p. 127)])

Fluoride is present in plaque and in dental calculus, and in the latter material it may be related to fluoride in the water.

EXCRETION OF FLUORIDE

Fluoride is excreted in the urine, sweat, and feces. It occurs in traces in milk, saliva, hair, and, presumably, tears. Fluoride is probably not exhaled in the breath, but definitive data are lacking.

The principal route of fluoride excretion is via the urine. The process is rapid, as illustrated in Figure 9-11. Several factors are recognized as influencing urinary excretion, such as total intake, the form in which fluoride is taken into the body, whether the person is relatively unexposed or regularly exposed to fluoride, and the person's general health, especially with regard to kidney disease.

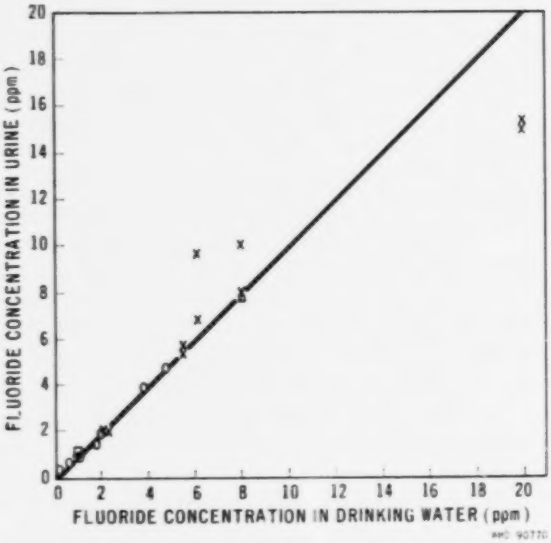

FIGURE 9-11 Relation between fluoride concentration in urine of humans and that in water supplies used. ○, McClure and Kinser.[421] X, Largent.[350] Δ, Likins *et al.*[369] □, +, Zipkin *et al.*[745] (Reprinted from WHO.[731] (p. 148))

Urinary Excretion

Measurement of the renal clearance of fluoride labeled with fluorine-18 in the human showed that fluoride clearance was greater than that of chloride, that fluoride clearance increased with urine flow, and that fluoride clearance was always less than creatinine clearance. Reabsorptions of 51% and 63% were observed in two subjects, compared with a chloride reabsorption of 99.5% or more. Less efficient reabsorption of fluoride accounts for the very high excretion rate.[123] Studies in the dog by Chen *et al.*[137] gave comparable results. It is concluded, therefore, that fluoride is removed from the circulation by glomerular filtration and that the rapidity of excretion results from the less efficient tubular reabsorption.

Urinary fluoride concentration is widely recognized as one of the best indices of fluoride intake. At least two differing circumstances of intake should be recognized.

In persons whose normal intake is fairly constant, the urinary fluoride concentrations may fluctuate as variable amounts are ingested in the usual mixed diet and as different amounts of water are drunk. Over periods of months, however, urinary excretion and skeletal concentrations tend to establish at least superficial steady states.

Some groups may undergo unusual exposure—for example, industrial exposure, high drinking-water concentrations, or excessive water consumption due to high environmental temperatures. Urinary excretion will be higher in these groups but will probably also reflect a steady state. However, persons who experience considerable but brief random exposures are "relatively" unexposed, in the sense that their skeletal tissues are by no means saturated. When the usual intake is temporarily grossly increased, the rapid processes of distribution and excretion deposit about half the additional fluoride in the skeleton and remove the remainder promptly to the urine.

The extraordinary relation between the concentration of fluoride in drinking water (up to 8 ppm) and the concentration in the urine is shown in Figure 9-11. Some scatter of concentrations among individuals does exist. In the United States, persons using water that contains little or no fluoride usually excrete urine with fluoride concentrations of 0.2–0.5 ppm. With a water supply containing 1 ppm, normal urinary concentrations often range from 0.5 to 1.5 ppm. The near equality of fluoride concentrations in the drinking water and urine is a striking feature of Figure 9-11, and this relation is a useful rule of thumb for predicting urinary fluoride concentrations (or concentrations in drinking water).

Urinary fluoride concentrations vary from hour to hour, day to day, and person to person. Figure 9-12 shows the day-to-day variation in fluoride excretion, expressed in milligrams per day and parts per million.[119] The values for subject B show a threefold variation. Subject A drank tea, which contains about 1 ppm of fluoride; as expected, he excreted more fluoride than did subject B, who was not a tea drinker.

The practical difficulties of collecting a 24-hr urine sample, compared with the relative ease of obtaining spot samples, raise the question of whether spot samples can be used. Comparisons of fluoride concentrations in 24-hr collections and in spot samples from the same persons during the same 24-hr interval have been reviewed.[287] One must conclude that 24-hr samples are more reliable and should

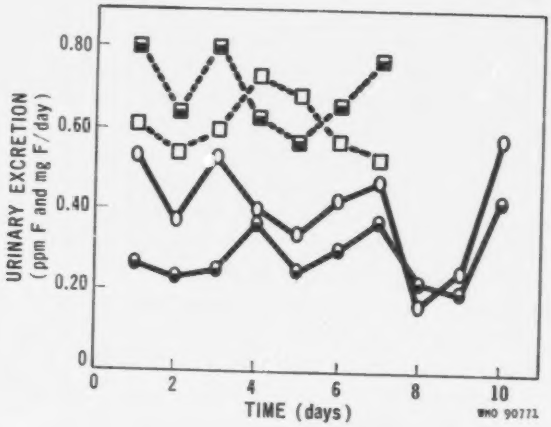

FIGURE 9-12 Day-to-day variations in urinary fluoride excretion. Data of Büttner *et al.*[119] ■- - -■, Subject A, mg/day. □- - -□, Subject A, ppm. ○—○, Subject B, ppm. ●—●, Subject B, mg/day. Subject A drank tea. (Reprinted from WHO.[731 (p. 149)])

be collected if the data for a single person are to form the basis for decision and interpretation. Fluoride analyses of spot samples, although they vary in the same person from time to time and in different persons at the same time in comparable exposures, give reasonably reliable averages for groups and can be used for industrial-hygiene control or population survey.

The rapidity of urinary excretion of fluoride has already been remarked on (*cf.* Figure 9-1); approximately 20% of ingested fluoride appears in the urine in about 3 hr.

The very high rate of excretion is one of the most important protective factors in severe fluoride poisoning—usually, the victim either dies within 4 hr or recovers. The critical period is short because fluoride is so rapidly removed from the circulation and extracellular fluid via the kidney and because skeletal deposition is also extremely rapid. Together, these processes effectively lower fluoride concentrations if the absorbed dose is not overwhelming.

In a relatively unexposed person, about half a single dose of fluoride is excreted in the urine within 24 hr, and about half is deposited in the skeleton. Although a number of experiments in humans and animals form the basis for this assertion, the most convincing evidence comes from the classic experiments of Largent and his colleagues. Over a period of several years, Largent measured the fluoride in samples of everything he ate, drank, and excreted, so that definitive measurements of fluoride retention were possible. Fluoride was ingested in daily doses of 1–18 mg, in the form of sodium fluoride or calcium fluoride in solution, as solid bone meal, as solid cryolite, and so on. Observation periods were one to several months long. A plot of Largent's data is shown in Figure 9-13, which clearly demonstrates that half the absorbed fluoride is removed from the system via the urine.

Younger persons, a greater proportion of whose skeletons is accessible to the circulation and who are actively laying down bone mineral, excrete a lower proportion of a given dose than do adults. For example, infants excrete 32–50% of the fluoride ingested daily.[75] Longwell[375] noted that children 5–6 years old excreted 0.16 mg/24 hr, whereas children 10–12 years old excreted 0.35 mg/24 hr. Gedalia[226] reported excretion by children 1–3 years old to be only half that of children 4–6 years old. The role of the skeleton in younger versus older persons is strikingly demonstrated in the study of Zipkin *et al.*,[745] in which urinary fluoride concentrations of children and adults were measured in a community before and after fluoridation of the community water supply. After fluoridation, fluoride concentration in the urine of adults increased from a prefluoridation value of about 0.3 ppm to 0.9–1.0 ppm, approximately that of the water supply (1 ppm), in about a week. The children, however, required

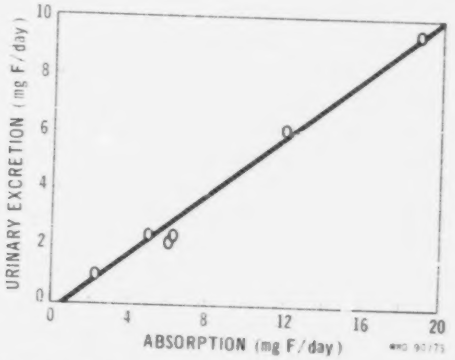

FIGURE 9-13 Relation between absorption and urinary excretion of fluoride. (Reprinted from WHO.[731 (p. 152)])

3 years of water-supply fluoridation for their urinary fluoride concentrations to reach 0.9 ppm.

Fluoride deposition in the skeleton of man is not irreversible. It has been observed that persons suffering from crippling fluorosis or osteosclerosis exhibit higher than normal urinary fluoride concentrations for long periods after the excessive fluoride exposure. Presumably, the process involves the loss of fluoride both from the surface and from the interior of the crystals of the bone mineral. If the plasma fluoride and extracellular fluoride concentrations decrease, exchangeable fluoride from the crystal surfaces is replaced by hydroxyl ions and freed to enter the circulation and be excreted. In the constant osteoblastic–osteoclastic reworking of the human skeleton, the osteons are destroyed, and mineral in the crystals thus dissolved is again available to be excreted or redeposited.

Hodge *et al.*[231 (pp. 158–159)] have examined Largent's data, describing fluoride intake and excretion, and plotted the rate of removal of the stored fluoride after Largent concluded his experiments on himself. The data are shown in Figure 9-14, from which it can be calculated that about 8 years would be required to remove half the fluoride stored in this person's skeleton.

A review of the available evidence by Hodge *et al.*[731 (pp. 155–156)] indicates that urinary excretion of fluoride tends to be reduced in persons with advanced kidney disease. In fact, Call *et al.*[121] found increased bone concentrations of fluoride in some but not all persons suffering from advanced chronic renal disease in their study group, and Taves *et al.*[666] have reported increased serum fluoride concentrations in patients with renal failure.

Urinary excretion of fluoride has been reported to be decreased during pregnancy and to return to prepregnancy levels 2–3 months after delivery.[227] Hodge *et al.*[731 (p. 157)] suggest that this may be the result of an increased capacity of the skeleton to take up fluoride as a result of hormone-induced bone changes before delivery.

In patients with primary osteoporosis or Paget's disease, urinary excretion of fluoride is normal or possibly decreased.[544]

Fecal Excretion

About 10% of the total daily fluoride excretion can usually be accounted for in the feces. A person ingesting the average diet in the United States and not using a fluoridated water supply usually excretes less than 0.2 mg/day in his feces. If the fluoride is ingested as

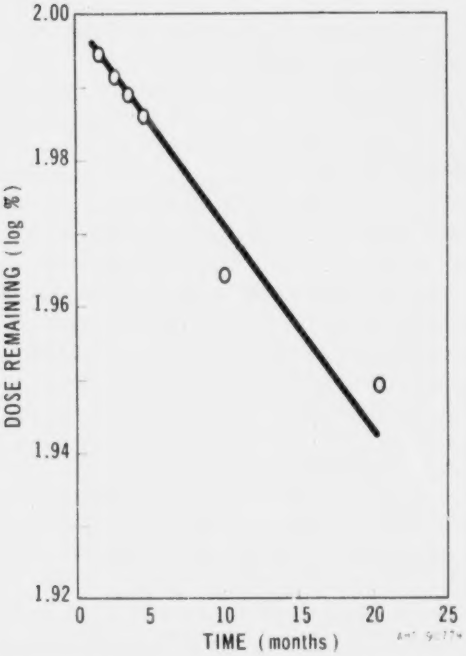

FIGURE 9-14 Mobilization of skeletal fluoride in man. Subject's skeleton was estimated to contain 1.7 g of fluoride retained from prior experimental periods of known fluoride ingestion.[352] The amounts mobilized during successive intervals after completion of the ingestion experiments were calculated from the reported urinary excretion data. (Reprinted from WHO.[731 (p. 158)])

relatively insoluble compounds—such as bone meal, cryolite, and calcium salts—or if such precipitants as aluminum and calcium compounds are present, larger proportions of the fluoride are unabsorbed in the intestinal tract and appear in the feces. This may amount to as much as 30% of that ingested.

With insoluble fluorides, the particle size of the ingested material becomes significant. For example, fluoride-containing rock phosphate powder ingested in a finely dispersed form, as may occur in a fertilizer plant, was recovered in the feces only to the extent of 13–19%.[350] Part of the fecal fluoride probably represents fluoride that has been absorbed and re-excreted via the gastric and intestinal juices. Re-excretion into the gut presumably accounts for the excess fecal fluoride observed after inhalation of hydrogen fluoride.[350]

Fluoride-balance data obtained in human subjects are shown in Table 9-14.

Excretion in Sweat, Milk, and Saliva

Some fluoride is lost from the body in sweat. Although there have been only a few investigations in this area, the available data indicate that during excessive sweating up to 50% of the total fluoride excreted may be lost via the perspiration. Rates may sometimes nearly equal those in urine.[731 (p. 143)] There is some evidence that the concentration in sweat increases when additional fluoride is ingested, but the significance of this route in normal conditions of sweating has not yet been established.

Only negligible amounts of the fluoride intake can be accounted for in milk and saliva.[193] The concentrations of fluoride in milk are approximately the same as those in blood; increased levels of fluoride intake are reflected in slight increases in the concentration in milk. Fluoride concentrations in saliva are low, and presently available data do not show a striking correlation with fluoride in drinking water.

ACUTE FLUORIDE INTOXICATION

After a large dose of a soluble fluoride is ingested the course of poisoning is violent and brief; deaths are frequently recorded in 2–4 hr. In the United States, deaths from fluoride poisoning (suicidal plus accidental) constitute approximately 1% of all the deaths by poisoning with all agents identified in *Vital Statistics of the United States*.[287]

The fatal dose of fluoride is usually unknown, but it has been recorded or deducted in a few cases. A dose range of 5–10 g of sodium fluoride can be cited as a "certainly lethal dose" for a 70-kg man; this corresponds to about 70–140 mg/kg. Gettler and Ellerbrook[23] estimated what they referred to as the smallest absorbed lethal dose of fluoride at 105 mg for a person who weighed about 65 kg. The figure was arrived at from chemical determinations of the fluoride in organs taken from five fatal cases. In reality, this represents only the

TABLE 9-14 Fluoride Balance in Humans[a]

Fluoride in Water Supply, ppm	Age, Years	Length of Residence, Years	Time of Observation, Days	Fluoride Ingested Daily, mg			Fluoride Excreted Daily, mg			Balance, mg/Day
				Fluid	Food	Total	Urine	Feces	Total	
2	35	10	96	2.4	1.2	3.6	2.9	0.4	3.3	+0.3
5.5	55	29	60	3.8	1.3	5.1	4.5	0.6	5.1	0
6.1	57	34	133	6.7	1.0	7.7	8.1	0.4	8.5	−0.8
8	57	19	140	11.3	2.5	13.8	10.4	1.4	11.8	+2.0
20	30	8	45	20.8	1.5	22.3	12.3	1.4	13.7	+8.6

[a] Adapted from WHO.[731] (p. 143)

fraction of the absorbed fluoride still remaining in the soft tissues at death; it might better be referred to as the "smallest soft-tissue burden" at the time of death from acute fluoride poisoning.

Tissue fluoride concentrations from those five cases and from a fatal poisoning by sodium fluorosilicate reported by Steenhauer and Kedde[625] are shown in Table 9-15. The data indicate an appreciable increase in the fluoride content of tissues as a result of the ingestion of lethal amounts of fluoride.

The clinical symptoms of acute fluoride poisoning and the frequency of their occurrence as recorded by Roholm[549] are listed in Table 9-16. After ingestion of large doses, there is diffuse abdominal pain; diarrhea and vomiting; excessive salivation, thirst, and perspiration; and painful spasms of the limbs.[731 (p. 227)]

The mechanism of fluoride poisoning has not been clearly established. Several features of this problem have been reviewed by Hodge and Smith.[287] Generally, fluoride kills in acute poisoning by blocking normal metabolism of cells. Enzymes involved in essential processes are inhibited. Vital functions, such as the origin and transmission of nerve impulses, cease. Functions controlled by calcium, such as blood clotting and membrane permeability, are affected. Cell damage and necrosis produce massive impairment of organ

TABLE 9-15 Fluoride Content (in ppm) of Human Tissues Taken at Autopsy[a]

	Fluoride Content[b]		
Tissue	Normal Deaths[c]	Death from Sodium Fluoride[c]	Death from Sodium Fluorosilicate[d]
Blood	0.20 - 0.64 (4)	3.5 - 15.5 (5)	5.8 (1)
Brain	0.40 - 0.68 (4)	1.6 - 3.4 (2)	–
Heart	0.44 - 0.60 (2)	10.6 (1)	–
Kidney	0.56 - 0.78 (2)	4.6 - 11.6 (3)	–
Liver	0.36 - 0.70 (3)	4.4 - 12.2 (5)	7.2 (1)
Lung	0.16 - 0.42 (3)	12.4 - 15.6 (2)	–
Spleen	0.25 - 0.30 (2)	11.8 (1)	–
Urine	0.40 - 0.56 (4)	–	68.8 (1)
Stomach contents	–	–	4.4 (1)
Small intestine contents	–	–	(in feces)

[a] Adapted from Hodge and Smith.[287]
[b] Parentheses enclose numbers of cases.
[c] Data from Gettler and Ellerbrook.[231]
[d] Data from Steenhauer and Kedde.[625]

TABLE 9-16 Frequency with Which Symptoms were Observed in 34 Fatal Cases of Acute Fluoride Poisoning[a]

Symptoms	No. Cases
Vomiting	31
Pain in abdomen	17
Diarrhea	13
Convulsions, spasms	11
General weakness and muscular weakness and collapse	8
Pain or paresthesia in extremities	7
Paresis, paralysis	5
Difficulty in speech and articulation	5
Thirst	5
Perspiration	5
Weak pulse	5
Change in facial color	5
Nausea	4
Unconsciousness	4
Salivation	3
Impaired swallowing	3
Motor restlessness	2
High temperature	2

[a] Adapted from WHO.[731(p. 227)]

function. Terminally, there is a characteristic shocklike syndrome. In dogs fatally poisoned with fluoride given intravenously, Caruso *et al.*[128] noted a fall in blood pressure and an increase in the rate and depth of respiration. The immediate cause of death was respiratory arrest; the heart continued to beat for a time after cessation of respiration. Terminally, ventricular fibrillation occurred.

Numerous instances of acute nonfatal poisoning have occurred. These are reviewed by Hodge and Smith.[287] The minimal symptomatic dose is approximately one fiftieth of the certainly fatal dose. Duration of illness ranges from one day to several weeks.

Several investigations of the effects of acute exposures to fluorine and hydrogen fluoride in animals have been published recently.[334,335,541,542]

CHRONIC EFFECTS OF FLUORIDE IN MAN

No instance of retarded human growth caused by the ingestion of fluoride is known. The size and development of children in the

American Southwest, where drinking-water supplies contain up to 5 ppm of fluoride, have never been questioned as abnormal. No distinction in developmental rates was found between children in Newburgh, N.Y., where the water was fluoridated to a concentration of 1 ppm, and children in Kingston, the control city.[287]

There has been some concern that increased intakes of fluoride may lead to an increased incidence of mongolism. Rapaport[533] recorded the number of mongoloid children in institutions in various cities as fractions of the numbers of births in the cities where the children were born. His data showed an apparent increase in mongolism with increases in the fluoride content of the water. Russell has pointed out, however, that, although the babies may have been delivered in hospitals in high-fluoride areas, many of the mothers resided in surrounding rural areas and did not use the community water supplies during their pregnancies. Therefore, the fluoride concentrations in the towns where the children were born are not necessarily those of the water used by the mothers, and Rapaport's statistics are therefore unacceptable. Furthermore, Russell was able to show that the differences described by Rapaport were not significant.[287] Using more carefully collected data, Russell compared the numbers of institutionalized mongoloids from the populations of areas using water containing no more than 0.9 ppm of fluoride with those from the populations of areas whose water supplies contained 1.5 ppm or more. The difference was so slight that it could have occurred 5 out of 6 times by chance.[287] Berry[72] compared the numbers of mongoloid children born with the total numbers of children born live to mothers from areas of England using water containing 0.7–2.0 ppm of fluoride and with those born to mothers from areas using water containing 0.2 ppm or less; areas using 0.2–4.0 ppm and less than 0.25 ppm were also compared. No relation was found between the fluoride content of the water supply and the incidence of mongolism.

During the last few years, a number of reports have appeared in which fluoride is alleged to have been the agent responsible for a variety of vague and variable symptoms, such as headache, nausea, vomiting, and epigastric pain. Most of these reports have come from a single investigator, G. L. Waldbott, who considers them to reflect an allergic response or an intolerance of fluoride. The problems associated with interpreting these reports have been reviewed by Jenkins[321] and by Schlesinger,[731 (pp. 309–310)] both of whom conclude that this condition, if it exists, has not yet been adequately documented.

Soft Tissue

There have been reports of histopathology in two extended series of autopsies of humans exposed to fluoride in the water supply or in the air. Call *et al.*[121] examined tissues from 88 persons known to have been exposed to increased concentrations of fluoride in the atmosphere and from 39 persons not meeting the authors' criteria for industrial exposure. No histologic changes that could be attributed to the fluoride exposure were seen. Geever *et al.*[229,230] autopsied 728 persons who had resided for from less than 5 years to over 20 years in Colorado Springs, where the drinking water contains 2.5 ppm of fluoride; no pathologic findings attributable to prolonged residence in this environment were found. Leone *et al.*[367] could find no differences in tissues of two persons of similar ages, one of whom had used an almost fluoride-free water for 25 years, and the other, water containing 8 ppm of fluoride for 34 years. Leone *et al.*[366] have also investigated the medical status of residents of a high- and a low-fluoride area. Although they did not study autopsy material, their report does detail the results of thorough physical examinations of the two populations, in relation to various organs and systems.

KIDNEY

Fluoride-induced renal pathology due to chronic exposure in man, with the causative agent established beyond reasonable doubt, has never been reported.[731 (p. 250)] Autopsy examinations have undoubtedly been conducted on tissues of persons with more than minimal fluoride exposure, but they have probably gone unrecognized in more instances than not. Few descriptions of findings have been published. However, the studies of Geever *et al.*,[229,230] Call *et al.*,[121] and Leone *et al.*[366,367] all call attention to the absence of changes in the kidney of man that can be attributed to fluoride.

No renal pathology ascribed to fluoride has been found in experimental animals maintained for long periods on diets or drinking water containing 50 ppm of fluoride or less. The borderline water concentration at which some individuals of some (not all) species exhibit changes is about 100 ppm.[287] The children in the Newburgh-Kingston study referred to earlier excreted similar amounts of albumin and sugar, whether the water was fluoridated or not.[568] The incidence of albuminuria and glycosuria in persons using a water supply containing 8 ppm of fluoride was not different from that in residents of an area using water with 0.4 ppm;[366] nor did McClure[420] find differences in the urine of young men using fluoridated water (2.0–5.2 ppm) and that of young men from low-fluoride areas.

THYROID

Two questions almost inevitably are raised regarding the thyroid and fluoride: Is fluoride accumulated in the thyroid as is another halide, iodide? Does absorbed fluoride interfere with thyroidal uptake of iodide and its conversion to thyroxine?

The answer to both questions is No. Over 30 years ago, Evans and Phillips[198] analyzed thyroid tissue of patients undergoing thyroidectomy. No correlation was found between the fluoride content of the thyroid, the iodide content of the thyroid, the basal metabolic rate, and the fluoride content of the patients' drinking water. More recent studies with fluorine-18 in normal persons and in hyperthyroid patients have shown no appreciable uptake of the isotope by the thyroid. After intravenous injection of fluorine-18 in mice and rats, no concentration of fluoride was found in the thyroid.[197] Several community studies in Switzerland, reviewed by Demole[731 (pp. 260–261)] have demonstrated that the addition of fluoride to a community water supply or to commercially available iodized salt has not interfered with the proven ability of iodide to eliminate and prevent thyroid hypertrophy in the population. The children of the Newburgh–Kingston study showed no effects of 1 ppm of fluoride in the water supply on the thyroid,[731 (pp. 305–308)] nor did 8 ppm of fluoride in the water significantly alter the thyroid gland in residents of Bartlett, Texas.[366] Animal experiments have demonstrated that histologic and functional changes can be produced by excessive doses of fluoride, but more than 50 ppm in the ration or drinking water was required to produce detectable or measurable changes.[287]

HEMATOPOIETIC SYSTEM

Various reports of hematologic changes in patients with fluorosis have appeared in the literature, but, as Singh and Jolly[731 (pp. 262–263)] point out, it is difficult to separate specific effects of fluoride from other factors, such as malnutrition and nutritional imbalance. Agate *et al.*,[17] in a survey of British industrial workers exposed to fluorides, found normal blood counts and hemoglobin concentrations. Macuch *et al.*[404] have noted some hematologic changes (e.g., decreased hemoglobin and increased number of erythrocytes) in children living near an aluminum-processing plant. No hematologic changes attributable to 1 ppm of fluoride in the drinking water were seen in the Newburgh–Kingston study.[731 (pp. 305-308)] According to Leone *et al.*,[366] the incidence of abnormal lymphocyte counts was greater in Bartlett, Texas, residents (8 ppm of fluoride in the drinking water) than in Cameron Texas, residents (0.4 ppm), but they did not consider that to be necessarily a fluoride effect. In general, the data from animal experiments indicate that there are changes in some but not all criteria at dietary levels greater than 100 ppm.[287]

HEART AND CARDIOVASCULAR SYSTEM

There is a paucity of data in the literature describing the effects of fluoride on the human heart and cardiovascular system.[287] Leone *et al.*,[366] in their survey of the health status of the populations of Bartlett (8 ppm of fluoride in the water supply) and of Cameron (0.4 ppm) noted a slightly higher rate of cardiovascular abnormalities in Cameron residents than in those of Bartlett. It is interesting that Bernstein *et al.*[70] found fewer calcified aortas, on x-ray examination, in persons using a water supply containing 4–5.8 ppm of fluoride than in persons whose drinking water contained only 0.15–0.3 ppm. Papoyan and Demirchoglyan[510] found x-ray evidence of hypertrophy of the ventricles in more than half the workers in the electrolytic shop of an aluminum plant, but electrocardiography revealed no damage to the myocardium. These investigators considered the changes seen to be a hypertrophy of the heart muscle comparable with that produced in the skeletal musculature, resulting from the constant physical work. An increase in capillary permeability was also noted in 60% of the workers; the origin and significance of the condition are not clear.

Several studies in animals have reported a decrease in blood pressure caused by fluoride. For example, Leone *et al.*[365] subjected dogs to intravenous infusion of fluoride at a dosage of 20 mg/kg, which resulted in a progressive depression of blood pressure and heart rate; 30.6 mg/kg led to a depression of respiratory rate and a conversion to atrioventricular nodal or ventricular rhythm with terminal ventricular fibrillation or asystole. Lu *et al.*[379] performed similar experiments, in which monkeys were subjected to fluoride perfusion to death. Atrial fibrillation was noted in one instance, and partial atrioventricular heart block in two others, shortly before death.

Caruso *et al.*[128] have studied the effects of fluoride on the heart in detail and have suggested that in the intact animal fluoride may produce a direct vasodilatory effect. The hypotensive response of dogs to oral doses of fluoride of 1 or 5 mg, as described by Richardson *et al.*,[546] could not be repeated by Caruso.

RESPIRATORY SYSTEM

Fluorine and hydrogen fluoride are pulmonary irritants that, in sufficiently high concentrations, can have devastating effects. Stokinger[634] has reviewed several studies of the effects of the two agents in dogs, rabbits, mice, and rats exposed to high concentrations for minutes or hours or to lower concentrations for 18-31 days, and Keplinger[334] and Keplinger and Suissa[335] have described more recent studies in laboratory animals exposed for 5-60 min to different concentrations of fluorine. The studies agree that the changes seen in the lungs include coagulation necrosis, congestion, ecchymotic or petechial hemorrhage, bronchitis, and bronchiectasis, the more severe effects being seen as a result of the more severe exposure conditions.

A measure of the incidence of respiratory effects of fluorides in man is available in several studies. Hough *et al.*[296] examined employees in steel mills where sodium fluoride is routinely added to molten steel and compared the findings with those obtained from steelworkers in a plant formerly using fluoride and from steel workers who had never used fluoride. The most commonly noted symptoms were cough and upper respiratory complaints, such as hoarseness, sore throat, and nasal congestion; cough and pharyngitis were noted less frequently among those exposed to fluoride than among those in the control plant. The pharyngitis may have resulted in part from the sodium fluoride, tar smoke, and sulfur oxide fumes at the furnace, but the effects of these three factors could not be separated. No pulmonary changes were related to sodium fluoride exposure.

Lyon[385] has reported on the incidence of respiratory complaints among employees exposed to gaseous fluorides and among employees in the same plant not so exposed. Such complaints occurred to a lesser extent in the fluoride-exposed workers; among the exposed workers, there was no apparent relation between the incidence and airborne concentrations of fluoride.

Derryberry *et al.*[177] determined the incidence of abnormal respiratory findings in the medical histories of persons working in a phosphate-fertilizer manufacturing plant and found it to be significantly greater in the medical histories of workers exposed to fluoride than in those of workers not exposed. However, on physical examination of the subjects, the difference in incidence no longer was significant, although it still was greater in the exposed workers. The weighted fluoride exposure of the men ranged from 0.50 to 8.32 mg/m^3, with a mean of 2.81 mg/m^3. According to Machle *et al.*,[386] the highest concentration of hydrogen fluoride that can be tolerated for more than 1 min is about 100 mg/m^3; respiratory and conjunctival irritation are marked. At 26 mg/m^3, there is discomfort, but the concentration can be tolerated for several minutes.

In essence, the available evidence summarized here indicates that, in conditions of good industrial hygiene, pulmonary and respiratory effects of fluoride exposure are of no serious magnitude.

SKIN

Several instances of dermatitis attributable to industrial exposures to fluorine, hydrogen fluoride, or sodium fluoride have been reported.[287] The "chizzola" maculae described by Waldbott[699] have not been seen in other investigations (N. L. Kaltreider, Aluminum Company of America, personal communication).

EYE

Concern has been expressed regarding possible effects of relatively high intakes of fluoride on the eye. For example, Geall and Beilin[225] reported a case of bilateral optic neuritis in a man receiving oral doses of fluoride for osteoporosis, and Waldbott[700] attributed a retinopathy in one patient and incipient changes in the retina of another patient to the use of fluoridated water. However, de Deuxchaisnes and Krane[171] found no abnormality in the ocular fundi and visual fields of five patients with Paget's disease who were being treated with sodium fluoride. Guerra and Medaglini[240] also reported no ocular changes in six patients receiving oral doses of fluoride for more than a year. More recently, Bloome and Burain[78] have investigated the effects of large dosages of fluoride as sodium fluoride (20 mg/kg–day) given orally to rabbits for 3–5 months, but found no significant differences in the electroretinograms, fundi, or microscopic sections of eyes between treated and control animals. Waterborne fluoride at 1 ppm did not produce ophthalmologic changes in the children of the Newburgh–Kingston study.[731 (pp. 305-308)]

NERVOUS SYSTEM

There are very few data available related to the effect of fluoride on the central nervous system (CNS). With regard to man, we are forced to fall back on data from studies of mortality statistics,[263,459] which merely indicate that ingested fluoride did not lead to an increase in deaths attributed to diseases of the CNS.

The situation is not much better with regard to animal studies. Lu *et al.*[380] reported that rats given sodium fluoride by intraperitoneal injection (10 mg/kg daily) for 4–14 weeks or 7 or 70 ppm of fluoride in the diet (which contained 50–60 ppm of fluoride chiefly as bone meal) for 15–19 weeks were more sensitive to chemical stimulation of the CNS by pentylenetetrazole or strychnine and to electric shock. The animals were also more sensitive to the CNS-depressant effect of diphenylhydantoin (reduction of incidence of electroshock) and showed a prolonged pentobarbital-induced sleeping time. The effect of fluoride on the activity of pentylenetetrazole and of pentobarbital disappeared 2–3 weeks after the animals were returned to a normal fluoride intake. F. A. Smith and L. C. K. Wong (unpublished data) attempted to reproduce the results of Lu *et al.* on pentobarbital sleeping time. They administered 8 and 63 ppm of fluoride in the drinking water to rats maintained for 8 months on a diet containing 18 ppm of fluoride, but the sleeping time was unaffected. Elliott[189] also was unable to confirm the observation of Lu *et al.*

Rice and Lu[543] showed that a diet of 70 ppm of sodium fluoride for 16 weeks caused an increase in sensitivity to the paralytic effects of succinylcholine in rats, which they attributed to the inhibition of cholinesterase activity. They also showed that rats receiving 1.68 or 3.36 mg of fluoride per 100 g of body weight daily for 30 days by stomach tube showed a heightened senstivity to the cholinesterase inhibitors, parathion and demeton.

Lu *et al.*[379] have also investigated the effect of intravenous lethal doses of sodium fluoride on the CNS of rhesus monkeys. The electroencephalogram changed from fast low-amplitude to slow high-amplitude activity. Depression and alternate periods of wakefulness and sleep were noted. Muscle fasciculations and clonic convulsions were occasionally seen. Pupils were of normal size and reactive to light until shortly before death, when they became dilated and fixed.

Apparently, only two reports in the literature have examined the behavioral effects of fluorides. Elliott[189] studied the effects of 150 ppm of sodium fluoride added to a standard laboratory diet on the ability of rats to learn a multiple T-maze. No differences were attributable to the added fluoride either in time taken to run the maze or in errors made while learning it. It should be pointed out, however, that there was no positive control group, so the sensitivity of the test is not known.

Sadilova[566] studied the much more toxic hydrogen fluoride. She reported on a series of conditioned-reflex studies in rats. Hydrogen fluoride was administered to the rats for 5 months in concentrations of 0.10, 0.03, and 0.01 mg/m^3. Her highest dosage produced an increase in the latency of some of the learned responses. She also found a disruption of some of the discriminations that the animals had been taught. These effects were greatest with the highest concentration but were detectable even with the 0.03-mg/m^3 dosage. Rats exposed to the latter concentration returned to control levels of performance after a 1-month recovery period. These behaviorally active dosages also were reported to have depressed cholinesterase activity. In addition, those exposed to the 0.10-mg/m^3 concentration showed, on histologic examination of the brains, changes ranging from hyperemia of the membrane capillaries to structural changes in the nerves themselves. Sadilova[566] also reported on human sensory thresholds to hydrogen fluoride. She estimated the minimal concentration for odor perception at 0.03–0.11 mg/m^3. About the same concentrations increased the sensitivity of the eye to light.

The presence of only those two studies makes any firm conclusions about behavioral effects of fluorides premature. In addition, the reader should keep in mind the absence of any data on the sensitivity of the maze test used by Elliott[189] in his negative study of sodium fluoride and the absence of confirmatory replications of the Sadilova[566] studies on hydrogen fluoride.

Bone

MECHANISM OF UPTAKE AND REMOVAL

In general, as the mineral content of bone increases, the fluoride content also increases. The amount of fluoride present depends on a number of factors, including fluoride intake, age, sex, specific bone, and specific part of the bone. The relation between amount of ash and amount of fluoride in the rat femur is plotted in Figure 9-15,

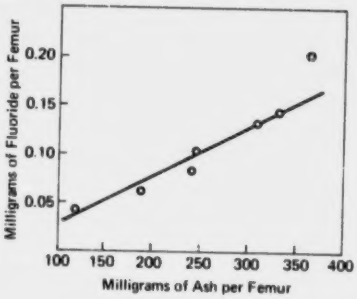

FIGURE 9-15 Increase of fluoride in rat femur as ash content increases. Data of Miller and Phillips.[456] (Reprinted with permission from Hodge and Smith.[287])

which indicates a constant increase in fluoride for each unit increase in femur ash—in this particular experiment, about 0.05 mg of fluoride for each 100 mg of ash. The fluoride : mineral ratio remained constant when the dietary fluoride intake was maintained during growth.

It has also been demonstrated that, as calcification increases, fluoride concentrations increase with increasing percentages of ash. The relation is shown in Figure 9-16, where it is also evident that fluoride in the femoral ash increases more rapidly in the female than in the male. The difference is tentatively ascribed to a larger available skeleton in the female.

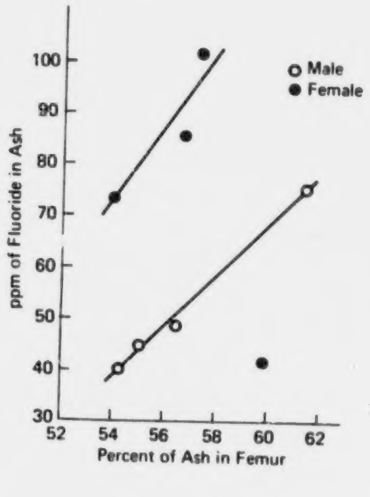

FIGURE 9-16 Increase in rat femur fluoride concentration as percentage of ash increases in the femur. Data of Bixler *et al.*[75] (Reprinted with permission from Hodge and Smith.[287])

Considerable evidence shows that the fluoride ion is able to replace the bicarbonate and the hydroxyl, but not the phosphate of bone salt. Neuman and Neuman[474] suggested that fluoride in extracellular fluid enters the apatite crystal by a three-stage heteroionic exchange process. The hydroxyapatite of bone mineral exists as extremely small crystals surrounded by a hydration shell. Fluoride first enters the hydration shell, in which the ions are in equilibrium with those of the surrounding tissue fluids and those of the apatite crystal surface. The second-stage reaction constitutes an exchange between the fluoride of the hydration shell and a monovalent group at the crystal surface. Once it has entered the surface of the crystal, fluoride is more firmly bound. In the third stage, some of the fluoride may migrate deeper into the crystal as a result of recrystallization. The consensus is that absorbed fluoride is incorporated into the hard tissues largely by the process of exchange and by incorporation into the apatite lattice during mineralization. Deposition by precipitation is probably minimal or absent.

Fluoride can be removed from bone, as is evidenced by its continued appearance in the urine in increased amounts after exposure or ingestion has ceased. Hodge[285] has suggested that such removal takes place in two phases—a rapid process taking weeks and probably involving an ionic exchange in the hydration shell, and a slower phase taking years and due to osteoclastic resorption of bone.

PHYSICAL STRUCTURE AND HISTOLOGY

The earliest descriptions of changes in physical structure and histology are those given bv Moller and Gudjonsson[465] and by Roholm.[549]

Geever *et al.*[2[illegible]] examined bone specimens and cartilage of persons who used water supplies containing up to 4 ppm of fluoride for at least 10 years before death. Specifically, evidence of focal calcification of the periosteum and adjacent tendons and fascia, osteoclasis, osteophytosis, and cortical thickness was sought, and marrow sections were examined for hematopoiesis and degree and extent of trabeculation. Incidental changes due to aging were noted in all groups, but there was no relation to fluoride intake. Weidmann *et al.*[708] also examined the ribs of persons drinking water containing less than 0.5, 0.8, and 1.9 ppm of fluoride and found no differences in resorption area of the trabeculae or of the compacta among the three groups.

Singh and Jolly[731 (pp. 240-242)] have described in detail the gross appearance of the skeleton of a person with advanced fluorosis who had lived in an area whose water supply contained 9.5 ppm of fluoride.

An increase in the crystallinity of bone apatite with increasing fluoride content has been reported by Zipkin *et al.*[749]

The specific gravity or density of fluorosed bone has been reported to be increased, decreased, and unaffected, as has its breaking strength.[287]

CHEMICAL CHANGES

Bone composition is relatively resistant to alteration by fluoride, and even dietary levels of fluoride that can produce illness when continued long enough have little effect on the chemical composition of the bone, other than an increase in fluoride concentration itself.

Zipkin *et al.*[747] analyzed iliac crests, ribs, and vertebrae of a number of persons who had used water supplies containing up to 4 ppm of fluoride for 10–87 years before coming to autopsy, with the results shown in Table 9-17. Calcium, phosphorus, and potassium in the bone ash were unaffected by mean fluoride concentrations as high as 8,000 ppm. Sodium decreased slightly as fluoride increased. Carbonate decreased about 10%, citrate decreased about 30%, and magnesium increased about 15% when the fluoride showed an eightfold increase.

FLUOROSIS

Skeletal changes in chronic fluorosis have been reviewed by several authors—e.g., Møller and Gudjonsson,[465] Roholm,[549] Hodge and Smith,[287] Weatherell,[704] and Singh and Jolly.[731] (pp. 240–242)

Crippling fluorosis resulting from industrial exposure to fluoride seldom (if ever) occurs today, owing to the establishment of and adherence to threshold limits for exposure of workers to fluoride. It has never been seen in the United States. A major characteristic of the cases described by Roholm is osteosclerosis affecting the entire skeletal system—the vertebral system and pelvis first and most severely. The relation between x-ray findings and length of employment in the cryolite industry, as summarized by Bosworth *et al.*,[89] is shown in Table 9-18.

Extensive production of exostotic bone is usually accompanied by increased resorption. In fluorosis in man, the balance of resorption and formation is in favor of formation.

TABLE 9-17 Concentrations of Constituents of Selected Human Bones as Related to Fluoride Concentration of Drinking Water[a]

Constituent	Percentage of Constituent[b] in Bones					
	Iliac crest		Rib		Vertebra	
	Low Fluoride[c]	High Fluoride[d]	Low Fluoride[c]	High Fluoride[d]	Low Fluoride[c]	High Fluoride[d]
Ash	53.5	58.6	56.0	57.4	49.8	52.3
F	0.08	0.69	0.08	0.70	0.10	0.80
Ca	38.8	38.4	38.8	38.4	37.8	37.8
P	17.5	17.7	17.5	17.7	17.5	17.5
Mg	0.50	0.57	0.50	0.60	0.50	0.61
Na	0.73	0.70	0.79	0.68	0.73	0.68
K	0.09	0.08	0.09	0.10	0.18	0.12
Carbonate	5.83	5.20	5.41	5.19	5.31	4.55
Citrate	2.23	1.56	1.92	1.39	1.95	1.30

[a] Adapted from WHO.[731] (p. 166)
[b] Ash expressed on a dry, fat-free basis. All other values are expressed on an ash basis.
[c] Fluoride content of drinking water, <1.0 ppm.
[d] Fluoride content of drinking water, 4.0 ppm.

TABLE 9-18 Radiographic Findings in Cryolite Workers[a]

Length of Employment, Years	Condition of Osseous System as Seen on X-Ray Examination
2.5	*1st phase* of osteosclerosis in pelvis and vertebral column; coarse and blurred trabeculae; diffuse increased density of bone
4.5	*2nd phase:* increased density and blurring of contours in pelvis and vertebral column, extended to ribs and extremities
11	*3rd phase:* greatly increased density of bone; irregular and blurred contours; all bones affected, particularly cancellous bones; extremities thickened; considerable calcification of ligaments of neck and vertebral column

[a]Adapted from Hodge and Smith.[287]

Chronic effects of fluoride on the skeletal system have been described in some geographic areas where drinking water contains excessive fluoride. This form of chronic fluorosis was first described in India, and has since been reported from Ceylon, China, South Africa, Japan, Saudi Arabia, the United States, Canada, and Europe.[731 (pp. 238–239)] The relation between osteosclerosis and the estimated fluoride ingested by long-term or lifetime residents is shown in Figure 9-17. Crippling fluorosis as seen by Roholm is estimated to result from the daily ingestion of 20–80 mg for 10–20 years.

Smith and Hodge[611] have offered the prediction that osteosclerosis as evidenced by x-ray examination will not be detected in bones containing about 4,000 ppm of fluoride (0.4% on a dry, fat-free basis).

It must be admitted that the mechanisms of the fluoride effects on bone are not well understood.[288] The primary effect of small doses of fluoride is the stimulation of the osteocyte (or ameloblast). Larger doses impair the function of the osteocyte and of the ameloblast. For instance, Johnson[287] has described mottled bone, which is analogous to mottled enamel. Fluoride in sufficient doses induces new bone formation with more osteoblasts than in normal bone; osteoclastic activity continues. With excess fluoride, the new bone is abnormal, as shown by its mottling, by its abnormal staining properties in undecalcified sections, and by its abnormal appearance

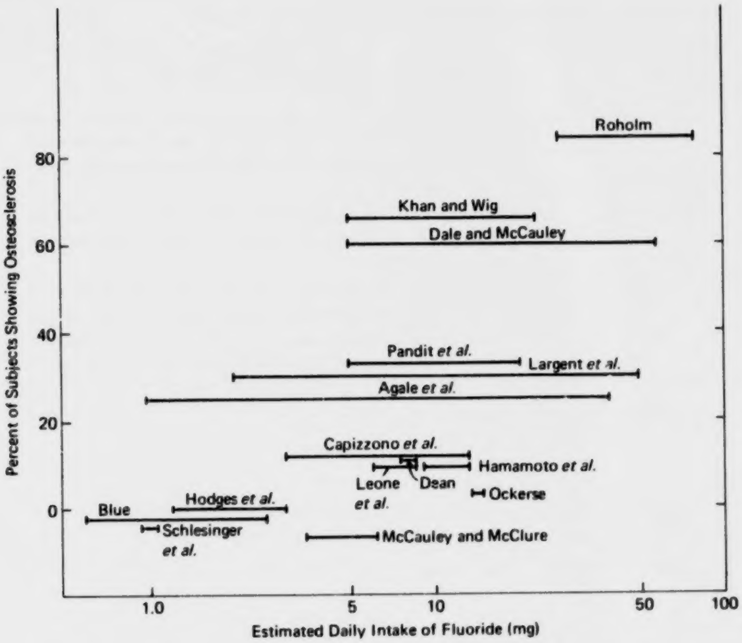

FIGURE 9-17 Relation of osteosclerosis to estimated ingested fluoride. (Reprinted with permission from Hodge and Smith.[287])

in polarized light, revealing collagen that is abnormal at least in alignment. In part, at least, the abnormalities in bone and collagen can be traced to abnormalities in matrix formation. The hypothesis that fluoride depresses bone-mineral solubility and tends to reduce serum calcium content sufficiently to stimulate the parathyroid gland to secrete parathyroid hormone and re-establish the normal serum calcium content is not yet confirmed.

Teeth

The uptake of fluoride by enamel and its relation to the amount of fluoride ingested have been described in an earlier section.

Dental tissues, like those of the skeleton, accumulate fluoride most rapidly during formation and mineralization. After growth and calcification are ended, the uptake of fluoride is almost entirely limited to tissue surfaces. In both dentin and enamel, the highest concentrations are found in marginal regions–at the pulpodentinal junction, where the blood supply is richest, and at the surface of enamel, which when unerupted has access to tissue fluid and when erupted is bathed in oral fluids.

During tooth formation, the cells of the dental tissues, particularly the ameloblasts, are very sensitive to fluoride. At relatively low doses–e.g., 2 ppm of fluoride in the water–small spots of discoloration may form in the tooth surface. These spots, or mottling, vary in color from paper white to dark brown. The brown stain of the latter condition usually accumulates after eruption. These doses affect only the appearance, and not the structure, of the tooth. At higher doses, the cells may be affected and the tooth structure severely altered, so that the normally smooth surface shows hypoplastic corrugations. These effects–mottled appearance and altered form–are produced only when the excessive amounts of fluoride are ingested during the period of formation and calcification, i.e., during the first 8 years of life in man. After the tooth erupts and calcification has been completed, ingested fluoride does not have adverse dental consequences.

RELATION OF BODY BURDEN TO DAILY INTAKE

Plasma

One might expect that a normal person in an apparent steady state with regard to fluoride exposure and intake would have a plasma fluoride concentration that would approximate the average concentration for all persons receiving the same fluoride input. This average concentration would be different for populations with different fluoride intakes. However, the extent to which this is the case is not clearly established. For example, Smith *et al.*[609] showed that the average blood fluoride content of persons whose water supplies contained 1 ppm of fluoride was only 3 times as great as that of persons whose water supply contained 0.06 ppm–a 17-fold difference in intake. Moreover, the variability of blood fluoride content in the samples was such as to make it impossible to determine from which population any single sample may have come. According to Singer and Armstrong,[600] plasma fluoride contents were indistinguishable among populations using 0.15-2.5 ppm of fluoride in the water, but were increased in persons whose water supply contained

5.4 ppm. They have suggested the presence of some homeostatic mechanism operating to keep plasma fluoride content reasonably constant.

Fluoride in blood has been measured on several occasions in personnel exposed to fluoride in industry. Pertinent data are shown in Table 9-19. A considerable discrepancy is apparent among the control data reported by the different authors. In three of the studies, no differences were found in the mean blood fluoride concentration of exposed and nonexposed persons. Wilkie[723] noted an increase by a factor of approximately 5 in one aluminum worker and an approximate doubling in a hydrogen fluoride worker. Smith and Gardner[287] also found an increase of blood fluoride by a factor of 1½ in fluoride workers.

The determination of fluoride in plasma is difficult, and the available evidence suggests that changes in plasma fluoride concentration, although important, may not be very large until fluoride intake has become relatively large. Consequently, the determination of plasma fluoride concentration is not recommended for the routine monitoring of fluoride intake.

Bone

The relation between fluoride intake and concentration of fluoride in bone has already been discussed. As the intake of fluoride is increased, either by increasing the dose or by increasing the time during which a constant amount of fluoride is ingested, the amount found in the bone also increases. At least 99% of the total body fluoride content is found in the calcified tissue (bone plus teeth).

Concentrations of fluoride in the bone of persons undergoing excessive exposure to fluoride have been reported in several instances, as shown in Table 9-20. As indicated previously, osteosclerosis detectable by x-ray examination is associated with fluoride concentrations greater than about 4,000 ppm in dry, fat-free bone.

Because of the difficulty or impossibility of obtaining routine biopsy samples of bone, measurement of bone fluoride cannot be recommended for monitoring fluoride exposure.

Urine

Attention has already been directed to the fact that urinary concentrations of fluoride are directly related to concentrations of absorbed fluoride.

TABLE 9-19 Mean Blood Fluoride Concentrations of Personnel Exposed to Fluorides in Industry[a]

Nonexposed Persons		Fluoride-Exposed Persons			
No.	Fluoride Content, µg/100 ml	Industry	No.	Fluoride Content µg/100 ml	Reference
24 (males)	255	Aluminum	77	213	Agate *et al.*[17]
–	47	Aluminum	1	299	Wilkie[722]
20	277	Magnesium	80	252	Bowler *et al.*[90]
–	–	Hydrogen fluoride	1	98	Wilkie[722]
12	12	Fluorides	12	30	F. A. Smith and D. E. Gardner (unpublished data)
–	230–300 (Avg., 270)	Hydrogen fluoride	5	200–400 (Avg., 270)	Gadaskina and Shtessel[213]

[a] Adapted from Hodge and Smith.[287]

TABLE 9-20 Bone Fluoride Concentrations in Persons Receiving Excessive Exposures to Fluoride[a]

Investigator	Bone	Age, Years	Fluoride, ppm in Ash	Normal Fluoride Content in Persons of Same Age, ppm
Linsman and McMurray[b]	Vertebra	22	7,500	450 ± 225
	Sternum		6,900	
Kilborn *et al.*[b]	Rib	37	20,830	690 ± 345
	Vertebra		18,650	670 ± 335
Wolff and Kerr[c]	Rib	48	15,900	850 ± 425
	Vertebra		19,880	830 ± 415
Roholm[d]	Rib	52	9,900	910 ± 455
	Vertebra		9,300	890 ± 445
	Rib	72	11,200	1,200 ± 600
	Vertebra		13,100	1,180 ± 590

[a] Adapted from Hodge and Smith.[386]
[b] Excessive fluoride in water.
[c] Rock phosphate dust.
[d] Cryolite dust.

Because of the ease of obtaining urine samples and the rapidity and ease with which the fluoride concentration can be determined with the fluoride-specific ion electrode, analysis of urine for fluoride is a recognized technique for surveying individuals or populations for fluoride exposure.

The long-term hazard to be protected against in the course of exposure to fluorides is the development of crippling fluorosis. If early osteosclerosis, as evidenced by increased density to x rays, can be avoided, then crippling fluorosis will never be seen. The question is: What concentration of urinary fluoride is associated with increased radiopacity?

As indicated earlier, a fluoride concentration of 6,000 ppm in dry, fat-free bone is associated with some osteosclerosis in approximately 10% of the population; osteosclerosis is not expected at a concentration of 4,000 ppm.[611] Zipkin *et al.*[748] have demonstrated that bone fluoride concentrations of 4,000 and 6,000 ppm are associated with water fluoride concentrations of 5 and 8 ppm, respectively, which in turn are associated with urinary concentrations of 5 and 8 ppm. On the basis of these data, osteosclerosis would not be expected in persons whose urine contained no more than 5 mg/liter (ppm).

Largent[350] has summarized some of the data from the literature pertaining to the relation between urinary fluoride and increased radiopacity of bone. He indicated that there was no increase in radiopacity in subjects whose urine contained 3.1–3.8 mg of fluoride per liter, but radiopacity was increased in subjects with higher fluoride content.

No changes in bone density were observed in workers chronically exposed to fluorides in an aluminum plant where the average urine concentrations in samples taken before the workshift varied from 1.6 to 3.6 ppm at a specific gravity of 1.024. The average taken immediately at the close of a work period of 3–5 days varied from 3.0 to 10.4 ppm, with 56% of the samples below 8 ppm at a specific gravity of 1.024 (N.L. Kaltreider, Aluminum Company of America, personal communication).

The Public Health Service, in 1966–1967, conducted a study of workers exposed to fluorides in a chemical plant where the mean air concentration of hydrogen fluoride was 1.03 mg/m^3, and that of particulate fluorides, 0.18 mg/m^3. The concentration of fluoride in the urine of 23 employees varied in preshift samples from 0.33 to 3.52 mg/liter (mean, 1.76), and in postshift samples, from 1.55 to 8.8 mg/liter (mean, 3.60). X-ray examination of seven workers with long-term or higher exposures to fluorides showed no evidence of skeletal fluorosis or deposition of fluoride. Ventilatory function tests on 305 workers were within normal limits for age in most of the men. Some depression in function found in a small percentage of the men did not appear to be work-related (M. M. Key, personal communication to A. M. Baetjer).

The experience of Derryberry *et al.*[177] and of Rye[564] suggests 4 mg of fluoride per liter as a reference level in determining the probability of increased bone density. Elkins[188] considers values above 3 mg/liter as indicative of excessive exposure.

Evidence from several sources thus indicates that urinary fluoride concentrations, in preshift samples, not exceeding 5 mg/liter are not associated with detectable osteosclerosis and that osteosclerosis is unlikely at urinary fluoride concentrations of 5–8 mg/liter.

SUMMARY

The human health effects associated with various levels of fluoride intake are summarized in Table 9-21.[286]

TABLE 9-21 Major Effects of Fluoride[a]

Effect	Dose	Time	Amount of Fluoride	Number of Times Amount Exceeds Probable Intake from Air at 10 ppb
Death[b]	Single	2–4 hr	2,500–5,000 mg	50,000–100,000
Kidney injury	Repeated	Months	>100 ppm[c]	2,500[d]
Thyroid injury	Repeated	Months or years	>50 ppm	1,200[d]
Body weight loss	Repeated	4 or more years	40 ppm[e]	1,000[d]
Crippling fluorosis[b]	Repeated	10–20 years	20–80 mg or more	500–2,000 or more
Mottled enamel[b]	Repeated	1st 8 years of life	2–8 ppm or more	50–200 or more

[a] Derived from Hodge and Smith.[387]
[b] Effect in man.
[c] In water.
[d] Assuming factors to translate animal exposure to human exposures.
[e] In feed.

As yet, no recommended daily allowance for fluoride has been put forth for humans, as has been the case with numerous other dietary components, such as vitamins, minerals, carbohydrate, protein, and fat. The closest approach to such a standard is the recommendation that fluoride-deficient water supplies be artificially fluoridated to a concentration of approximately 1 mg/liter (or somewhat less for warmer climates, where more water will be ingested), to reduce the incidence of dental caries.

The daily intake of fluoride consists of approximately 0.2 mg from foods, some amount contributed from drinking water (which depends on the concentration of fluoride in the water supply), and fluoride from industrial sources, dentifrices, and possibly drugs. For the average U.S. adult not working in a fluoride industry and using a water supply containing 1 mg/liter, the approximate daily intake is 1.2–1.5 mg of fluoride. There are no data available describing the total body burden of fluoride in persons who have ingested that much daily. The only data that can be offered are those of Smith,[606] who, with Gardner, measured fluoride in the total ash of a series of exsanguinated human cadavers (Table 9-22). Residential histories of these subjects are not available, but at least a large portion of their lives was spent in western New York, where the water supplies generally contain not more than about 0.1 ppm of fluoride. In this

TABLE 9-22 Fluoride Content of Ash of Human Cadavers[a]

Age, Years	Sex	Total Ash Wt., kg	Fluoride in Ash, ppm	Total Fluoride, g
Fetal	–	–	140	–
Fetal	–	–	29	–
36	M	2.48	918	2.27
48	M	4.41	744	3.28
54	M	2.50	659	1.65
55	M	2.50	1,480	3.70
57	M	2.10	2,270	4.76
60	M	2.50	550	1.37
65	M	3.35	828	2.78
66	M	1.97	1,006	1.98
68	M	2.50	1,020	2.55
74	M	2.26	1,485	3.36
74	M	2.31	717	1.66
75	M	2.00	1,187	2.39
76	M	2.00	1,144	2.29
77	M	1.70	2,145	3.65
77	F	0.79	933	0.74
81	M	1.57	988	1.55
82	M	2.58	1,494	3.86
83	M	2.20	1,506	3.31
85	F	1.50	567	0.85
85	M	2.57	1,288	3.31

[a] Bodies had been used for instruction purposes; samples of ash made available through kindness of Dr. C. E. Tobin, Department of Anatomy, University of Rochester School of Medicine and Dentistry, Rochester, N.Y. Derived from Smith.[606]

limited series, there appears to be no relation between fluoride content and total ash weight. The age span of the subjects is limited, and no relation is evident between fluoride concentrations and age. The data do indicate that total fluoride in the human body is appreciable—0.74–4.76 g, with an average of 2.57 g in this series. This is comparable with the total body stores of iron or zinc and appreciably more than the stores of manganese, copper, or iodine.

Urinary concentrations of fluoride in man are directly related to the levels of absorbed fluoride. Urinary concentrations in relation to associated air concentrations of fluoride are listed in Table 9-23. In most instances, the exposures reported were chiefly exposures to gaseous fluorine compounds. The relation between urinary fluoride content and atmospheric fluoride exposure is not well established. The best data available for such a comparison are those of Collings

TABLE 9-23 Concentrations of Fluoride in Air and in Urine

Industry	Air Fluoride Concentration	Urine Fluoride Concentration, mg/liter	Reference
Brazing	0.24 mg/m^3	3.1	Elkins[188]
Silver soldering	0.17 mg/m^3	2.1	Elkins[188]
Welding	0.47 mg/m^3	2.3	Elkins[188]
Magnesium melting	0.48 mg/m^3	3.9	Elkins[188]
Not specified	0–2.4 mg/m^3	1.0 preshift 3.2 end of shift	Collings *et al.*[147]
	2.5–4.9 mg/m^3	0.8–3.7 preshift 5.1–10.4 end of shift	
	5.0–9.9 mg/m^3	1.2–3.9 preshift 5.8–10.1 end of shift	
	>10 mg/m^3	3.5–5.6 preshift 10.0–19.0 end of shift	
Phosphate processing	6.35–7.70 ppm	6.47	Poppe[526]
	1.18–2.77 ppm	7.86	
	8.8 ppm	7.35	
	14.6 ppm	4.08	
	5.90 ppm	2.90	
	1.54–30.90 ppm	2.86	
	5.90–25.0 ppm	3.67	
	19.8 ppm	2.89	
	1.78–7.73 ppm	0.5–44.0	Derryberry *et al.*[177]
	0.5–8.32 ppm	0.2–43.0	
	≤4 ppm	up to 5	Rye[564]
Magnesium foundries			
Core shop	0.143 mg/m^3	0.9–4.1	Bowler *et al.*[90]
Foundry	0.286–6.37 mg/m^3	0.5–7.5	
Furnace	0.314 mg/m^3	1.0–3.4	
Uranium processing; gaseous diffusion	0.8 ppm	1.1	Lyon[385]

et al.,[146] who reported data for airborne fluorides in industry and for before-shift and after-shift urine samples. Largely on the basis of those data, Elkins[188] concluded that an airborne fluoride concentration of 2 mg/m^3 should correspond approximately to 5 mg/liter of urine if the sample is obtained at the end of the shift. Hodge and Smith[286] arrived at a comparable estimation: allowable daily intake of 5–6 mg of fluoride for a worker exposed to the threshold limit value of particulate fluoride, i.e., 2.5 mg/m^3.

Unfortunately, many studies for a better evaluation of the effects of airborne fluoride on human health remain to be done. Not many authors have investigated the incidence and magnitude of effects on the thyroid gland, the hematopoietic system, the cardiovascular system, and the central nervous system. However, these systems respond readily to a number of stresses, not only to fluoride, and a causal relation to airborne fluoride has been established only poorly or not at all. More careful studies are required, with better attention being paid to the nature of the responses, the presence or absence of other medical or physical conditions that might contribute to the occurrence of the responses, and the proper control groups.

The airborne fluorides to which subjects are exposed must be better evaluated with respect to amounts of fluoride-containing material, proportions of gaseous and particulate fractions, chemical and physical properties (including particle size) of the particulate fraction, and meteorologic conditions in the surrounding community when resident populations are being studied.

Meanwhile, we must rely on the evidence that airborne fluoride concentrations that produce no plant response contribute quantities of fluoride that are negligible in terms of possible adverse effect on human health and offer a satisfactory margin of protection for man.

10

Population Hazards

POPULATION STUDIES

Analysis of mortality data from communities with different amounts of fluoride in the water supply can provide indirect evidence about the effects of ingested fluoride on physiologic systems and organs. The approach has been used by several investigators.

Hagan[245] has summarized three such studies carried out in the United States. The first of these, by the Illinois State Department of Public Health, compared rates of mortality from all causes and from heart disease, cancer, nephritis, and diabetes among Illinois communities using water supplies containing 0.8 ppm of fluoride or more and 0.4 ppm or less.[304] It was concluded that there was "little or no support for claims of adverse effects being produced by limited ingestion of fluorides." In the second study, 32 pairs of cities in different parts of the country were selected. Of each pair, one city used a water supply containing 0.7 ppm of fluoride or more, and the other city used a water supply containing 0.25 ppm or less. The 1950 rates of these cities for deaths from all causes, from heart disease, from cancer, from intracranial lesions, from cirrhosis of the liver, and from nephritis showed no differences. The third analysis was based on mortality experience in five U.S. cities before and after the addition of fluoride to the local water supply. The mortality experience in 1940 was compared with that in 1950 (before and after 1–5 years of fluoridation), after suitable adjustments for population changes. For each of the cities, a comparison was made of the recorded deaths and the expected deaths calculated from age–sex–race mortality data for the United States as a whole. A significant change in this ratio from year to year would have indicated the effect of some factor other than age, sex, and race. However, for none of the five cities did the 1950 ratio of recorded to expected deaths differ significantly from that for 1940.

Heasman and Martin[263] compared mortality in natural fluoride areas (0.4–5.8 ppm of fluoride) with that in areas deficient in fluoride (less than 0.2 ppm) in England and Wales, in much the same manner as did Hagan *et al.*[245,246] The list of causes of death was extended, however, and included all causes, tuberculosis, all cancer, cancer of the stomach, cancer of the lung, leukemia, vascular lesions of the central nervous system, angina, other heart diseases, influenza, pneumonia, bronchitis, peptic ulcer, nephritis and nephrosis, congenital malformations, all other diseases, accidents, poisonings, and violence. Some significant differences were observed when individual causes of death were compared, but even when the higher incidence occurred in residents of higher-fluoride areas, the figures were below the national or regional averages. Heasman and Martin considered it very unlikely that fluoride was the cause.

A similar study of population statistics was reported from Italy 2 years later by Mirisola and Cruciani.[459] Data from districts using water containing 1–3 ppm of fluoride (Lazio region, near Rome) were compared with those of fluoride-deficient areas, one of which was newly settled and therefore had a nonindigenous population. The following causes of death were considered: infectious and parasitic diseases; malignant tumors; mental illnesses and those of the nervous system, including vascular lesions of the central nervous system; diseases of the circulatory system (acute articular rheumatism and diseases of the heart and blood vessels); respiratory diseases; digestive diseases (ulcers, liver); other diseases (allergic, endocrine, metabolic, nutritional, and urinary tract diseases, cogenital malformations and diseases of early infancy); senility; and unidentified. There were no significant differences in average age at death among residents of the three areas. Tabulation of cause of death and average

age at death showed some differences. For deaths caused by cancer, diseases of the nervous system (including mental illness), and circulatory, respiratory, and other general diseases, the age of death was higher in the fluoride area; for deaths due to infectious diseases and senility, the age was lower in the fluoride area. Also comparison of the average age at death and the incidence of specific diseases (including infectious diseases and tumors) with corresponding data for Italy as a whole showed no significant differences.

There was a lack of evidence of a fluoride effect on rheumatic complaints in a population whose water supply had been artificially fluoridated to a concentration of 1 ppm for 5 years.[32] The prevalence of rheumatic disease was no greater in this population than in persons residing in an area where the water contained only negligible amounts of fluoride.

A survey by Bernstein *et al.*[70] showed a lower prevalence of osteoporosis and collapsed vertebrae in persons whose water supplies contained 4–5.8 ppm of fluoride than in persons using water containing 0.15–0.3 ppm.

Such studies as these have their limitations, particularly as regards the crude nature of mortality data and the selection of comparable populations. Nevertheless, these investigations are unanimous in their failure to find any effect that could be clearly attributable to fluoride.

SUSCEPTIBILITY OF SPECIAL POPULATION GROUPS

Some attention has been given to the question of unusual susceptibility of children to fluoride. The obvious effects of fluoride occur in children, whose teeth are being formed; these include the desirable effects of fluoride (reduction in dental caries) and the undesirable (mottling of teeth), and it has been well established that the difference is due to the amount of fluoride absorption.

Some investigations of the sensitivity of children to fluoride have focused attention on the nondental effects of fluoride and on the effects of airborne fluoride. If airborne fluoride were to cause an effect, the effect could be direct, through inhalation of airborne material, or indirect, through deposition of airborne fluoride on food and water that are later ingested. Agate *et al.*[17] examined adults and schoolchildren living near Fort William, Scotland, aluminum factory, which discharged large amounts of fluorides to the environment; they also studied factory workers. Physical, radiographic, blood, urine, and dental examinations were performed. Children living in a village about a mile from the factory had a significantly lower incidence of caries than found in other Scottish children, but the children in Fort William, about 1.5 miles from the factory, did not. The water supply of both villages was low in fluoride (70–180 ppb). The reason for this difference in caries incidence was not found. There were no abnormal x-ray findings in the children from the two villages. Blood counts (red-cell white-cell, and differential counts) were within normal limits for both groups of children, but the data were not compared with those of a control population. Other than a few cases of mottled teeth and the possible effect on caries incidence discussed above, these children were not affected by their exposure to fluoride by the criteria examined.

Demole *et al.*[172] studied groups of children of the same ethnic background living in different parts of Switzerland, some in areas of high soil and water fluoride content, and others in areas of low fluoride content. They found a lower incidence of dental caries in areas where the water contained 1.4 ppm of fluoride than in another area where the water contained 0.05 ppm. Children drinking the water with the higher fluoride content had no significant thyroid changes. Other changes possibly attributable to fluoride ingestion were not looked for.

Antonelli[33] examined workers from a fluoride-discharging industrial plant and children living nearby. Like most other investigators, he found a lower incidence of dental caries in children exposed to fluoride.

Savara and associates[567] found no signs or symptoms of fluoride changes in children living on Sauvie Island, Oregon. This island is near an aluminum reduction plant, whose discharge of fluorides was blamed for plant damage. Much of the food eaten by Sauvie Island families was homegrown, but drinking water came from wells whose water contained less than 0.1 ppm of fluoride.

A very careful study of children drinking fluoridated water over a 10-year period was conducted by Schlesinger *et al.*[568,569] The cities of Newburgh and Kingston, about 50 miles apart on the west bank of the Hudson River in New York State, were selected because of their similarity in size and in demographic, social, and economic characteristics. They had similar water supplies, deficient in fluoride (about

0.05 ppm), but the Newburgh water supply was fluoridated to a concentration of 1.0–1.2 ppm. Thorough studies of urine and blood, x-ray examinations, and ophthalmologic and otologic examinations were performed over a period of 10 years. No significant differences in the health of these children (other than differences in dental health) were found. There were no significant differences in hemoglobin and blood counts, but in the fluoridated city, there were a few more children with hemoglobin concentrations below 12.9 g/100 ml and red-cell counts below 4,400,000/ml, whereas there were a few more children in the low-fluoride area with white-cell counts above 10,000/ml. There was a significantly higher rate of tonsillectomies among children in the fluoridated area; it was attributed to differences in medical practice in the two cities and prevailed at the initiation of fluoridation, as well as during the study.

Sadilova[565] studied the teeth of children living near aluminum plants that discharged sufficient fluoride to damage plants and etch windows. There was generally a higher incidence of mottled teeth and a lower incidence of caries in these children than in children in a control village. The fluoride content of the drinking water in at least some of the villages was too low to account for the mottling of teeth. The fluoride content of food was not reported. Air concentrations of fluoride as high as 0.6 mg/m^3 were reported near one plant; this maximum was recorded within 0.5 km of the plant.

Balazova and co-workers[51] examined children from a village near an aluminum plant in Slovakia and found a lower incidence of dental caries in them than in children in a control village. These workers also reported reduced hemoglobin and increased erythrocyte counts in children from the affected village, compared with children from a control village, but no changes in total leukocyte counts and differential, x-ray changes in bones, or other signs or symptoms were observed. The criterion of significance of differences in these blood counts was statistical; the changes in red-cell counts do not seem to be of physiologic significance. Furthermore, the blood counts of the children from the control village suggest a mild anemia, which makes it more difficult to draw valid conclusions from the data.

Lindberg[371] studied the effects of discharges from a superphosphate plant on children's health. The control population was from a district 10 km from the superphosphate plant and had no nearby industrial establishments, and air analyses confirmed the greater cleanliness of the air in the district. The air near the superphosphate plant was high in fluoride (0.5 mg/m^3), sulfur dioxide, sulfuric acid aerosols, and oxides of nitrogen. Children exposed in this area, compared with the control population, had a higher incidence of catarrhal signs and symptoms; an increased frequency of enlarged lymph glands, especially of the neck; and a higher incidence of both tuberculous and nontuberculous chest x-ray changes. It seems unlikely that fluoride in the air was responsible for these changes, but a contribution by fluoride cannot be ruled out. There was a mild degree of dental fluorosis in the children living near the plant,[372] but no cases of dental fluorosis in the control area. The incidence of dental caries was 48–58% in the exposed area and 80% in the control area. The fluoride content of the water was low in each area. The fluoride content of food was not reported.

Macuch and co-workers[404] have also reported mottled teeth and a lower incidence of dental caries, as well as anemia, in children who lived near an aluminum plant in Czechoslovakia and were thus exposed to fluorides.

Kvartovkina *et al.*[347] studied children living near an aluminum plant that discharged fluorides into the atmosphere. They found greater standing and sitting heights in 5- to 7-year-old girls, but no other changes in physical measurements. They examined blood morphology and biochemistry and the phagocytic reaction of blood neutrophils and found a few changes, some of them seasonal. There was an increased morbidity from upper and lower respiratory diseases in younger children living near the factory, but without changes in white-cell counts, except for eosinophilia and a decreased leukocytic phagocytosis in winter. Older children showed some decreases in red-cell counts and hemoglobin concentrations, fasting blood sugar, and phagocytic reaction, together with eonsinophilia.

The reports of anemia in children do not seem adequate to draw conclusions on the hematologic effects of fluorides. See Chapter 9 for a fuller discussion of hematologic effects in persons absorbing fluorides.

Persons with advanced kidney disease seem not to be able to clear fluoride adequately from the blood into the urine, so more bone fluoride is deposited in such persons. Pregnant women excrete more fluoride than others. Mongolism has also been attributed to fluoride, but this has not been substantiated. Unsupported claims of an allergic response to fluoride have also been made. For a fuller discussion of these points, see Chapter 9.

In summary, the only established changes in children exposed to fluorides are in the teeth, and they include mottling and decreased occurrence of caries. Inasmuch as fluorides affect the teeth primarily during their development, the effects of fluoride on teeth would be expected to occur only in children.

STANDARDS FOR OCCUPATIONAL EXPOSURES TO FLUORIDES

Air-quality standards for work areas have been developed by individual industries, by technical associations, and by government agencies. These standards have been given various names, such as "threshold limit values," "maximum allowable concentrations," and "maximal acceptable concentrations"; the last two are commonly referred to as "MAC's."

In the United States, the most commonly used and accepted occupational health standards are the threshold limit values (TLV's), published yearly by the American Conference of Governmental Industrial Hygienists (ACGIH), and the U.S.A. standard acceptable concentrations (formerly called "maximal acceptable concentrations"), published by the United States of American Standards Institute (formerly the American Standards Association). Some states promulgate their own standards, but these usually follow such nationally and internationally recognized standards as TLV's and U.S.A. standard acceptable concentrations.

The standards referred to are for an 8-hr day, or 40-hr work week, over a man's working lifetime. They apply to healthy working adults, and not to elements of the population that might be especially sensitive to a specific chemical substance. (It is assumed that infants, elderly persons, and those with various diseases are more sensitive to chemical intoxication, although the validity of this assumption is variable.) Exposure for a normal work week allows periods of overnight and weekend recovery from transient effects and allows time for excretion of absorbed compounds and their metabolites. For these reasons, occupational-health standards will often be higher than those for air pollution in a community, where exposure is limited neither to healthy adults nor to a specific work schedule.

These occupational-health standards are designed to prevent adverse effects of long, repeated, low-level exposure to toxic substances. In the case of most inorganic fluorine-containing substances, the adverse effect that adherence to the standards should prevent is deposition of fluoride in bones in amounts that will cause the development of skeletal fluorosis. In the case of highly reactive materials like gaseous fluorine and hydrogen fluoride, the adverse effect to be prevented is a local effect on the respiratory system.

The TLV of fluorine gas is 0.1 ppm* (about 0.2 mg/m^3 †).[25] These values are based on experimental work of Stokinger,[634] in which animals tolerated 30-day exposures at 0.5 ppm but not at 2 ppm or higher. The ACGIH[26] believes that exposure of men at or below 0.1 ppm will be safe. The recommended maximal atmospheric concentration of the American Industrial Hygiene Association[27] is also 0.1 ppm.

Both the ACGIH,[25,26] and the U.S.A. Standards Institute[688] accept the safety of hydrogen fluoride at 3 ppm (approximately equivalent, in fluoride content, to TLV of fluoride, 2.5 mg/m^3). Exposure above that concentration is expected to cause a mild redness of the skin without significant discomfort. The recommended maximal atmospheric concentration of the American Industrial Hygiene Association[28] is also 3 ppm. The U.S.A. Standards Institute recommends that all hydrogen fluoride concentrations in air be kept below 10 ppm. But the ACGIH,[26] citing a report of Elkins[188] that nosebleeds occurred in workers at 0.4–0.7 mg/m^3, recommended that 3 ppm be a "ceiling" limit—that is, that the concentration of hydrogen fluoride be kept at or below 3 ppm.

The ACGIH[25,26] recommended a TLV of fluoride of 2.5 mg/m^3, on the basis of studies of animals and man by Ronzani,[555] Roholm,[549,551] Irwin,[307] and Largent.[351] Irwin's experience was that workers exposed at over 4 mg/m^3 excreted urinary fluoride at an average rate of 6.3 mg/day and developed a small incidence of slight radiographic bone changes, whereas exposure at 3–4 mg/m^3 resulted in no significant amount of bone changes. The ACGIH[26] concluded that the limit of 2.5 mg/m^3 "is sufficiently low to protect against permanent disabling bone changes." The U.S.A. Standards Institute,[688] which recommends 2.5 mg/m^3 as an acceptable concentration, reports that beginning changes of fluoride deposition in bone have not been found to be injurious in any way, so that chronic injury can be prevented by following skeletal x-rays. Excessive absorption of fluorides will be quickly reflected in increased urinary fluorides. The report suggests that fluoride absorption reflected in urinary fluoride concentrations consistently at or above 8 mg/liter (about 8 mg/day if the common assumption of daily urinary excretion of 1 liter is accepted) will result eventually in increased osseous radiopacity, whereas fluoride absorption reflected in urinary excretion of 4 mg/liter will not result in fluoride-induced radiopacity. The recommended maximal atmospheric concentration of the American Industrial Hygiene Association[27] is also 2.5 mg/m^3.*

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*Volumes of airborne gas or vapor per million volumes of air at 25 C and 760 torr.

†Milligrams of airborne substance, either gaseous or particulate, per cubic meter of air.

Smyth[615] has critically reviewed TLV's and the evidence on which they are based and concluded that the 2.5-mg/m^3 TLV of fluoride "is not low enough to prevent fluoride storage with resulting effects on bones." However, it should be noted that bone storage of fluoride occurs even with very small daily intakes of fluoride–for example, from dietary fluoride–and that even slight changes in bone demonstrated by x-rays have not been found to be injurious. It appears that daily occupational exposure to fluoride at 2.5 mg/m^3 is safe in healthy adult workmen. In any case, this occupational standard would not be expected to be low enough for an air pollution standard.

Only a few countries believe that 8-hr daily industrial exposure at an average concentration of 2.5 mg/m^3 is too high and recommend 1.0 mg/m^3 instead. Among these are the Soviet Union, Poland, Hungary, Czechoslovakia, and East Germany.[163] West Germany, however, recommends 2.5 mg/m^3.

A governmental air-quality standard for fluoride in the ambient air of inhabitated areas has been established only in the Soviet Union and several other eastern countries in the form of a maximal allowable concentration for atmospheric pollutants.[457,563] Their levels–e.g., 0.01 mg of fluoride per cubic meter for 24-hr average concentration with a maximal allowable single peak concentration of 0.03 mg/m^3–are much lower than any occupational health standard and questionable in their practicality. According to Ryazanov,[561,562] they are based on the presumption that the limits of allowable pollutant concentrations in the ambient air must be considerably lower than those recommended for the atmosphere in industrial plants. The workmen are exposed to the harmful effects of air pollutants in the plants for only 8 hr at a time, whereas the residents of inhabitated regions breathe the polluted air constantly.

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*See Chapter 9 for additional discussion of relationship between urinary fluoride levels and fluorosis in humans.

From a practical point of view, these limits constitute more a final goal still to be achieved than a true reflection of an existing situation in the vicinities of fluoride emission sources, even in the countries where they were established.

Threshold limit values have been developed in the United States for many other fluoride-containing substances (see Table 10-1). Most of the compounds for which TLV's have been recommended do not present air pollution problems, and many do not even present workroom problems, in that in many cases industrial exposure concentrations are significantly below TLV's.

TABLE 10-1 Threshold Limit Values of Various Fluorine Compounds

Fluoride-Containing Substance[a]	ACGIH Threshold Limit Value ppm	ACGIH Threshold Limit Value mg/m^3	Pennsylvania Short-Term Limit
Boron trifluoride (C)	1	3	1 ppm for 5 min
Bromine pentafluoride	0.1	0.7	–
Chlorine trifluoride (C)	0.1	0.4	0.1 ppm for 5 min
Dichlorodifluoromethane	1,000	4,950	–
Dichloromonofluoromethane	1,000	4,200	–
Dichlorotetrafluoroethane	1,000	7,000	–
Difluorodibromomethane	100	860	–
Fluoride (as F^-)	–	2.5	10 mg/m^3 for 30 min
Fluorine	0.1	0.2	0.5 ppm for 5 min
Fluorotrichloromethane	1,000	5,600	–
Hydrogen fluoride (C)	3	2	3 ppm for 15 min
Nitrogen trifluoride	10	29	–
Oxygen difluoride	0.05	0.1	–
Perchloryl fluoride	3	13.5	–
Selenium hexafluoride	0.05	0.4	–
Sodium fluoroacetate (1080)	–	0.05	–
Sulfur hexafluoride	1,000	6,000	–
Sulfur pentafluoride	0.025	0.25	–
Sulfuryl fluoride	5	20	–
Tellurium hexafluoride	0.02	0.2	–
1,1,1,2-Tetrachloro-2,2-difluoroethane	500	4,170	–
1,1,2,2-Tetrachloro-1,2-difluoroethane	500	4,170	–
1,1,2-Trichloro-1,2,2-trifluoroethane	1,000	7,600	–
Trifluoromonobromomethane	1,000	6,100	–

[a] C = ceiling value–i.e., a value not to be exceeded even briefly.

The toxic effects to be prevented by adherence to these standards vary among compounds. Several examples of different types of toxic action (or lack of toxic action) are given by fluorides. Some fluorohydrocarbons, used as propellants or refrigerants, are not metabolized to a form resulting in significant bone deposition; they have a low toxicity and a high TLV. Some compounds have a high toxicity for reasons other than their ability to cause fluorosis—for example, the highly toxic disulfur decafluoride (S_2F_{10}), commonly called sulfur pentafluoride, which causes pulmonary edema. Perchloryl fluoride is an example of another type of toxic action; it causes methemoglobinemia in experimental animals and is metabolized to release fluoride, which is deposited in bones; its TLV of 13.5 mg/m^3 is equivalent in fluoride content to the fluoride TLV, 2.5 mg/m^3.

Threshold limit values are time-weighted average concentrations for an 8-hr day, with reasonable excursions above the TLV permitted so long as the average is at or below the TLV. In the case of some substances, the ACGIH believes that excursions above the TLV should not be permitted and recommends "ceiling" values. This leaves inadequate guidance on what constitutes a "reasonable excursion" and on situations in which only brief exposures occur in work, rather than fairly steady concentrations over a working day. To help correct these deficiencies, the Commonwealth of Pennsylvania Division of Occupational Health recommends short-term limits for some substances.[148] These limits are recommended concentrations of airborne substances that can be safely inhaled for the stated periods (see Lieben[368] for a discussion of Pennsylvania limits). Pennsylvania short-term limits for fluorine-containing compounds are included in Table 10-1.

11

Summary and Conclusions

SOURCES AND CONCENTRATIONS OF FLUORIDE IN AMBIENT AIR

Fluorides are widely distributed in the earth's crust, in combined form, in gases, such as hydrogen fluoride and silicon tetrafluoride, and in particulate substances, such as cryolite, fluorapatite, calcium fluoride, and aluminum fluoride. Airborne fluorides are derived from natural sources, from industrial processes, and from fluoride-containing materials used by the public. Once fluorides have entered the atmosphere, their dispersal and their continued suspension in or loss from the atmosphere are determined not only by meteorologic conditions but also by the chemical stability and physical state of the fluorides and by their interaction with water vapor in the atmosphere.

The normal concentration of fluorides in the ambient air is low. Of a large number of air samples analyzed, 97% from nonurban areas had no detectable fluorides, and the highest concentration was 0.16 $\mu g/m^3$. Of samples from urban areas, 87% had less than 0.05 $\mu g/m^3$, and the concentration ranged from 0.05 to 1.89 $\mu g/m^3$ in the others. Higher levels occurred in areas close to industries that emit fluorides in their processes.

ENVIRONMENTAL ANALYSES

Sensitive, accurate, and precise methods of sampling and analysis for fluorides in air, water, soil, and plant and animal tissues have been developed. Atmospheric fluoride can be collected and separated into gaseous and particulate fractions by several methods.

FLUORIDE INTAKE, STORAGE, AND EXCRETION IN MAN

In general, the daily intake of fluoride inhaled from the ambient air is only a few hundredths of a milligram—a very small fraction of the total fluoride intake in man. For instance, if a person is exposed to ambient air containing about 8 μg of fluoride per cubic meter, his total daily intake from this source is calculated to be approximately 150 μg. Such a daily intake of fluoride might not even be detectable. It has been estimated that fluoride intake from other sources—water, food, therapeutic agents, and so on—usually approximates 1,200 μg/day, including about 1,000 μg from water (the amount usually recommended in temperate zones to reduce the incidence of dental caries) and about 200 μg from food and other sources.

Gaseous fluorine and hydrogen fluoride are absorbed from the respiratory tract and through the skin, and ingested soluble fluorides are absorbed through the gastrointestinal tract. Absorbed fluoride is excreted in the urine, and unabsorbed fluroide, in the feces.

Fluoride is present in plasma in concentrations of a few hundredths of a microgram per milliliter. Fluoride retained in the body is found almost entirely in bones and teeth, with less than 10 μg/g (fresh weight) in the soft tissues. Under normal conditions, atmospheric fluoride represents only a very small portion of the body fluoride burden.

EFFECTS OF FLUORIDE ON MAN

Severe effects of airborne fluoride on man, such as crippling fluorosis, have so far been observed only from long-term occupational exposures. Crippling fluorosis resulting from industrial exposure to fluorides, as once seen in the Danish cryolite industry, has never been seen in the United States. Osteosclerosis, as evidenced by increased density to x rays, may be associated with a concentration of about

6,000 ppm of fluoride in bone (dry, fat-free basis). Analysis of urinary fluoride is recommended as a means of monitoring populations for possible excessive exposures to fluoride; osteosclerosis is not expected in persons whose urinary concentrations of fluoride do not exceed 5 mg/liter.

Only a few instances of health effects in man have been attributed to community airborne fluoride, and they occurred in investigations of the health of persons living in the vicinity of fluoride-emitting industry. Most of these reports addressed themselves to the incidence of nonspecific responses, such as hematologic changes and other indices of general state of health, which are not peculiar to fluoride and may reflect various other influences. In fact, such effects have not been reported in the United States, even in persons exposed occupationally to airborne fluoride if the concentrations have not exceeded the recommended industrial threshold limit values. The current TLV for fluorine gas is 0.1 ppm v/v; for hydrogen fluoride, 3 ppm; and for particulate fluoride (as F^-), 2.5 mg/m^3.

The other responses attributed to fluorides in man have been found mainly in relation to nonairborne fluorides. The most sensitive response seems to be the production of mottled enamel in developing permanent dentition. Other effects, predicted from animal studies, may include loss of body weight and altered structure and function of thyroid gland and kidney; such effects have been reported to coincide with ingestion of excessive amounts of fluoride in food and water by animals.

EFFECTS OF FLUORIDE ON ANIMALS

Animals are exposed to fluoride from the air, ingested fresh forage or hay, feed supplements, and the water supply. Inhalation contributes only a negligible amount to the total fluoride intake of animals. In areas in which fluoride pollution is a problem, high-fluoride vegetation is the major source of fluoride in the ration of livestock.

The ingestion of fluoride by animals causes a number of specific lesions. Developing teeth are extremely sensitive to fluoride, and the subsequent abnormalities in permanent teeth are the most obvious sign of fluoride ingestion. Fluoride ingestion can also cause osseous lesions, lameness, and an appetite impairment that can result in decreased weight gain or diminished milk yield. Chemical evidence of the accumulation of fluoride in skeletal tissues, urine, and, to some extent, soft tissues can be used to monitor ingestion of fluoride by animals.

The available evidence indicates that dairy cattle are the most sensitive domestic animals, and it has been concluded that protection of dairy cattle from adverse effects will protect other classes of livestock.

Controlled experiments have indicated that long-term ingestion of fluoride in a concentration greater than 40 ppm in the ration of dairy cattle is needed to produce a significant incidence of lameness, any effect on growth or milk production, severe osseous lesions, or a significant incidence of severe dental fluorosis. Continual ingestion of a ration containing less than 40 ppm will result in chemical evidence of fluoride ingestion, discernible but nondamaging bone lesions, and changes in dentition that do not affect wearing quality. Inasmuch as developing dentition is also sensitive to short-term exposures to high concentrations of fluoride, there is no assurance that limiting the fluoride intake to a specific yearly average will protect cattle from severe dental fluorosis. Therefore, if full protection is to be ensured, limits must be placed on the period during which higher intakes can be tolerated.

On the basis of available experimental data, it has been suggested that dairy cattle can tolerate the ingestion of forage that averages 40 ppm for a year and that the average fluoride concentration of forage should not exceed 60 ppm for more than two consecutive months or 80 ppm for more than one month. The fluoride concentration of the total ration will in most cases be less than these values, because the animals will also be ingesting a feed supplement containing a lower fluoride concentration. Feeder cattle and lambs, which are exposed to fluoride for shorter periods, can tolerate higher but as yet undetermined concentrations of fluoride.

EFFECTS OF FLUORIDE ON VEGETATION

The ambient air is the primary source of phytotoxic fluoride, although smaller amounts are contributed by the soil and water. The more phytotoxic forms of airborne fluorides are hydrogen fluoride and silicon tetrafluoride gases and the soluble particulate substances, such as sodium fluoride and ammonium fluoride. Fluorides from the atmosphere are absorbed predominantly by the foliar tissues, although some may pass into the soil and be absorbed through the roots.

Accumulation of atmospheric fluorides by plants can result in changes in metabolism, production of foliar lesions, and alteration in growth, development, and yield. Plants may be grouped in three general classes, according to their response to fluoride exposure: susceptible, intermediate, and resistant. In addition to differences among species and varieties, the duration of exposure, stage of development and rate of growth, rate of accumulation of fluoride, environmental conditions, and agricultural practices are important factors in determining the susceptibility of plants to fluorides.

The following threshold concentrations for atmospheric fluorides are based primarily on research, rather than on field studies.

For exposure periods of 1 day, the threshold for foliar markings is between 3 and 4 $\mu g/m^3$ for the most susceptible species and 10 $\mu g/m^3$ or higher for species of intermediate susceptibility; for exposure periods longer than a month, the threshold is about 0.5 $\mu g/m^3$ for susceptible and between 1 and 3 $\mu g/m^3$ for some intermediate species. The relation of the accumulated foliar fluoride to the occurrence of foliar lesions is complex. Susceptible plant species show foliar lesions when tissue fluoride concentrations are 20–150 ppm. Some highly resistant species can tolerate tissue concentrations in excess of 4,000 ppm without injury. Reduction in growth may occur at tissue fluoride concentrations of 30–300 ppm, depending on the species and conditions.

The average concentration of fluoride in forage that appears to be important for animals is 40 ppm. Estimates suggest that a fluoride accumulation of 40 ppm in forage would be achieved in 30 days at a mean hydrogen fluoride concentration in the air of 0.33–1.3 $\mu g/m^3$. However, these estimates do not take into account variability in exposure to fluoride, rate of plant growth, or the effects of weathering in a field condition.

The available data suggest that a threshold for significant foliar necrosis on susceptible species or an accumulation of fluoride in forage of more than 40 ppm would be a 30-day average air concentration of gaseous fluoride of about 0.5 $\mu g/m^3$.

SUMMARY TABLE

To compare the relative levels of fluoride that are important for vegetation, cattle, and human subjects, the pertinent data have been combined in Table 11-1.

TABLE 11-1 Summary of the Effects of Fluoride on Vegetation, Cattle, and Human Subjects

Mean Concentrations					
Hydrogen Fluoride in Air, mg/m^3	Total Fluoride in Ration, Diet, or Water, mg/kg	Mean Daily Intake of Fluoride, mg/day	Duration of Exposure	Species	Effect Reported
0.00025	–	–	7 weeks	Chinese apricot	Less than 1% necrosis on foliage
<0.0005	–	–	18 months	Citrus	No effects on growth or physiology of plant
0.0006	–	–	1 week	Sorghum	Threshold for foliar markings
≈0.0007	–	–	2 weeks	Sorghum	Foliar lesions and reduced growth if exposed during anthesis
0.0009	–	–	17 weeks	Orchard grass and alfalfa	No effect on yield, no foliar lesions
0.0011	–	–	7 weeks	Chinese apricot	Some necrosis on foliage
0.0015	–	–	1 day	Gladiolus	Less than 2% necrosis
			1 day	Ponderosa pine	Less than 1% necrosis on young needles
			1 week	Sweet corn	Threshold for foliar markings
0.0018	–	–	2 weeks	Apple	Threshold for foliar lesions
0.0021	–	–	10 weeks	Bean	Significant effect on reproduction, but no foliar lesions
0.0029	–	–	22 weeks	Tomato	No effect on growth nor injury to foliage
0.0050	–	–	1 day	Gladiolus	About 5% necrosis of leaves
0.0063	–	–	2 weeks	Tomato	No injury
0.010	–	–	1 day	Tomato and some tree fruits	Threshold for foliar lesions

≃0.1	–	–	Single exposure	Man	Perceptible odor concentration for hydrogen fluoride
2.5	–	–	Daily	Man	8-hr daily exposures ACGIH threshold limit value for hydrogen fluoride
≃25.0	–	–	Single exposure	Man	Tolerable, with conjunctival and respiratory discomfort only for several minutes
≃100.0	–	–	Single exposure	Man	Highest tolerable concentration for 1 min, marked conjunctival and respiratory irritation
	–	0.2–1.2	Daily	Man	Average daily intake from American diet and water supply
	2–8[a]	–	Years	Man	Mottled enamel[b]
	20–30	–	Several years	Cattle	Threshold of discernible dental mottling[b]
	30–40	–	Several years	Cattle	Evident dental mottling[b]
	40–50	–	Several years	Cattle	Threshold of enamel hypoplasia,[b] moderate periosteal hypertosis
	>50	–	Several years	Cattle	Decreased milk production, significant incidence of lameness
	40[d]	–	Several years	Man[e]	Loss of body weight
	>50[a]	–	Months to years	Man[e]	Thyroid injury
	>100[a]	–	Months	Man[e]	Renal injury
	–	20–80	10–20 years	Man	Crippling fluorosis
	–	2,500–5,000	Single dose	Man	Death in 2–4 hr

[a] In water.
[b] Dental effects seen only if fluoride is ingested during period of tooth formation.
[c] Based on feeding experiments lasting 1.6–8 years.
[d] In diet.
[e] Based on data from animals extrapolated to man.

GENERAL EVALUATION

It is evident that the more important aspect of fluoride in the ambient air is its effect on vegetation and its accumulation in forage in amounts that lead to harmful effects in cattle and other animals. A comparison of current levels of fluorides in the community atmosphere with the concentrations required for injury to vegetation and cattle suggests that the hazard to these receptors is limited to particular areas.

Current knowledge indicates that airborne fluoride presents no direct hazard to man, except in industrial exposure. However, through the commercial, aesthetic, and ecologic functions of plants, fluoride in the environment may indirectly influence man's health and wellbeing.

12

Recommendations for Future Research

- Methods for sampling and separating gaseous and particulate forms of airborne fluoride have been developed and tested in various laboratories. There is an urgent need to subject methods, especially those to be recommended as methods of choice for collection and analysis of airborne fluoride, to collaborative and comparative studies among laboratories and in various field conditions. The Halogen Sub-Committee of the Intersociety Committee on Methods for Ambient Air Sampling and Analysis might be invited to function as an organizing body and arbitrator in performing these studies.
- The gross effects of fluoride on plants and animals have been studied and documented, but little is known about the basic biochemical lesions that are induced by fluoride. An expanded effort in this area is needed to furnish a complete understanding of the toxicity of inorganic fluoride.
- Fluorocarbon aerosol propellants have been produced and used in rapidly increasing amounts in the United States in recent years. The amount of fluorine released to the atmosphere in this form was estimated to be 45,000 tons in the United States in 1968. However, the distribution of these fluorine-containing compounds in the

environment, and the effects of the compounds and their degradation products on man, animals, and vegetation have not been adequately studied. This situation should be reviewed in light of an increasing and more widespread use of these compounds. Greater use should be made of the instrumental methods of analysis in determining their fate and effects; they can be detected at very low concentrations by modern instrumental methods of analysis, such as gas chromatography, mass spectrometry, and infrared and Raman spectrometry, but they pass undetected through most chemical analyzers.

- There is insufficient information on the general relation between the concentrations of atmospheric fluorides and their accumulation in forage plants. Controlled studies on the mechanisms and rates of accumulation and internal and superficial contamination of plants by different forms of atmospheric fluoride in field conditions are needed.
- Because dairy cattle are the most sensitive domestic animals and controlled experiments have shown the importance of time and fluoride concentrations in protecting foraging animals, experimental testing should be carried out in field conditions to establish the validity, in various foraging domestic animals, of any forage standard that might be recommended for the protection of livestock against the effects of increased fluoride intake.
- Industrial populations afford an important source of information on the effects of prolonged exposure to airborne fluorides in man. Although a few industrial studies have been made, more data of this type are greatly needed. Studies of industrial groups exposed to airborne fluorides should consider: concentration and form of fluorides in the working environment; fluoride concentration in urine samples collected at various times in relation to exposure; periodic radiology of bone tissue; respiratory function; complete blood counts and multiphasic screening; and careful physical examination, together with analysis of medical records and other suitable procedures to determine the general health pattern and any specific conditions that might be related to the exposures. Other sources of fluoride intake should be determined, where possible, such as concentrations of fluoride in the ambient air and water supply, so that total intake can be estimated for both the industrially exposed group and a comparable but nonexposed control group.
- Concentrations of fluoride in the ambient air that are safe for vegetation and animals are recognized to be low enough to prevent

the appearance of most of the effects of fluorides on human health. However, the possibility should be investigated that there are additive or synergistic interactions between fluorides and other airborne contaminants–such as ozone, sulfur dioxide, molybdenum, vanadium, and manganese–that could alter the long-term effects of low atmospheric concentrations of human health, animals, and vegetation.

- General health conditions of persons residing in the vicinity of fluoride-emitting industry have not yet been investigated as thoroughly and as carefully as they should be. More frequent and more extensive studies–e.g., of the nature of responses and of the presence or absence of other medical or physical conditions that might contribute to the occurrence of the responses–are required. Any relation to anemia should be explored. Special attention must be devoted to the establishment of proper control groups. The airborne fluorides to which the subjects are exposed must be evaluated better with respect to amounts of fluoride-containing material, proportions of gaseous and particulate fractions, chemical and physical properties (including particle-size analysis) of the particulate forms, and meteorologic conditions in the surrounding community when resident populations are being studied. The investigators should be aware that some persons in these populations–e.g., those with pulmonary and renal dysfunctions–might be especially susceptible to fluoride.

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